JOURNAL OF ECONOMIC ENTOMOLOGY

ENTOMOLOGICAL SOCIETY OF AMERICA

Vol. 51 August, 1958 No. 4

Warehouse Fumigation of Flue-Cured Tobacco with HCN to Control the Cigarette Beetle[1]

DANA P. CHILDS,[2] *Stored-Tobacco Insects Laboratory,*[3] *Richmond, Virginia*

ABSTRACT

Distribution of HCN in tobacco warehouse fumigation was studied when the gas was released by different methods. Uniform distribution was obtained in 1 hour when the gas was released throughout the entire warehouse; 4 hours required when it was released along the central aisle.

When a 16-ounce-per-1000-cubic-foot dosage of HCN was used, gas concentration was low throughout the entire fumigation. A booster shot of an additional 16 ounces after 8 hours greatly increased the rate of recovery. After 4 hours, the difference in recovery from 24- and 32-ounce dosages was 2 ounces or less. There was little difference in recovery from 32-ounce dosages either released all at once or half released as a booster shot.

Dosages of 16 and 24 ounces per 1000 cubic feet gave high mortality of cigarette beetle (*Lasioderma serricorne* (F.)) larvae to a depth of 3 inches in tobacco. Thirty-two ounce dosages were effective to 5 inches. As shown by weekly trap records, the 32-ounce dosage was more effective than a 24-ounce dosage, and the 24-ounce more effective than a 16-ounce dosage.

Very often warehouse fumigation is the most economical, practical, and efficient method of insect control for large quantities of tobacco. However, poor results are sometimes obtained with fumigation. Some unsatisfactory results may be partially attributed to control recommendations which have changed very little in the past 20 years. Methods of handling and storing tobacco during the same period have changed appreciably. The studies of this report were designed to determine: (1) the distribution of HCN throughout a warehouse equipped with a permanently installed piping system, as compared with a common procedure in which temporary rubber hose lines are used for introducing HCN; and (2) concentration and effect of fumigant when using varying dosages and booster shots to improve the efficacy of the fumigation.

In the spring of 1957, the Flue-Cured Tobacco Cooperative Stabilization Corporation had over 600,000,000 pounds of tobacco in storage held as collateral for Government loans. The tobacco was from crop years 1952–56, and thus much of it had ample time to become infested with the principal stored-tobacco pest, the cigarette beetle, *Lasioderma serricorne* (F).

For many years, timing of tobacco warehouse fumigation has been determined by the weekly catches of insects taken in suction-light traps. In Flue-Cured Tobacco Stabilization Corporation warehouses, usually, when the weekly trap catch of either the cigarette beetle, or the tobacco moth (*Ephestia elutella* (Hbn.)) reached a peak of brood emergence of 2000 or more, a fumigation was scheduled. Advantage of that opportunity was taken to conduct research on fumigation under industrial conditions. These data would not have been obtainable without the cooperation of the Flue-Cured Tobacco Stabilization Corporation and Fumigators Incorporated.

MATERIALS AND METHODS.—The warehouses used in these tests were constructed of lock-joint sheet-metal siding, with concrete floors and composition roofs. A central aisle divided each warehouse into two sections. The two warehouses used for comparison of a piped and unpiped section were each of 806,400 cubic foot capacity, and each was filled with approximately 5940 hogsheads of tobacco. The warehouses used for dosage and distribution studies were approximately 500,000 cubic feet in volume, each filled with approximately 4000 hogsheads of tobacco.

In the warehouses not permanently piped, the HCN was released through nozzles attached to rubber hose lines laid on the floor of the central aisle. In the piped warehouse, in addition to the lines laid on the floor, HCN was also released overhead through a permanently installed copper tubing system. The piping system traversed the length of the building, and approximately halved each of the sections on either side of the central aisle. Air pressure of 90 p.s.i. was used to force the HCN into the warehouses. In all tests the fumigation was for 72 hours.

Gas samples for determining the concentration of the fumigant were drawn through plastic tubing from predetermined locations throughout the warehouses at approximately 1-, 4-, 8-, 12-, 48-, and 72-hour intervals after

[1] Accepted for publication December 12, 1957.

[2] The author desires to acknowledge the assistance of Mr. Arthur F. Press in one of these experiments and express appreciation to Joseph N. Tenhet for advice and suggestions in planning and carrying out these studies.

[3] This is one of the field stations of the Stored-Product Insects Section, Biological Sciences Branch, Marketing Research Division, Agricultural Marketing Service, U. S. Department of Agriculture.

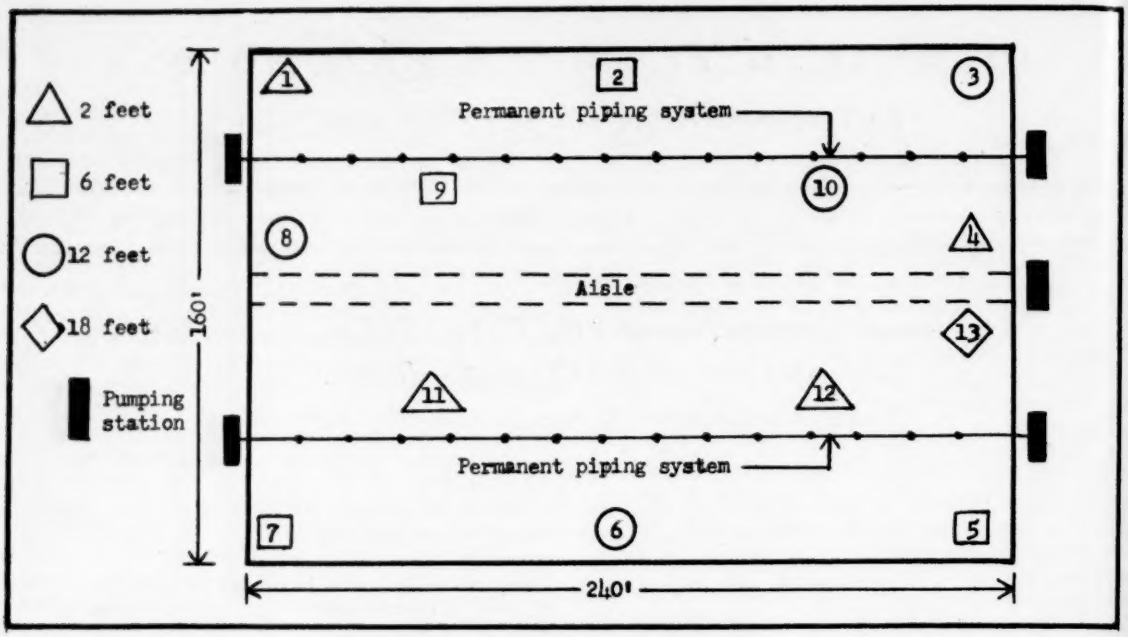


FIG. 1.—Floor plan of tobacco warehouse showing permanently installed piping system, pumping stations, and location of sampling points for gas concentration studies.

the end of the release of the fumigant. The technique used for HCN analysis was to trap the HCN in an alkaline solution, in previously evacuated quart bottles, and subsequent analysis by the standard Liebig, silver nitrate, titration method.

Simultaneously, biossay data were obtained by exposing approximately 3-week old cigarette beetle larvae in hogsheads of tobacco. This was accomplished by using insect test spikes designed to place lots of 25 larvae at each of 1-, 3-, 5-, 7-, and 9-inch depths in the tobacco. The spikes, systematically distributed throughout the warehouse remained in the tobacco for the 72-hour fumigation, and for an additional 72-hour post-fumigation. Larval mortality was recorded 24 hours after the removal of the spikes from the tobacco.

The success of a fumigation was determined from the follow-up of weekly trap catches of insects in the storage warehouses. Previous to the release of the gas, all suction-light insect traps were emptied and disconnected. Operation of the traps was resumed at the end of the 72-hour fumigation, and weekly records were taken for the remainder of the experiment.

DISTRIBUTION OF HCN.—Previous data on the efficiency of HCN fumigations in large warehouses had left some doubt as to whether the fumigant was uniformly distributed throughout the warehouse when released only from nozzles distributed along a central aisle. To obtain data on that point, 13 sampling areas, as shown in figure 1, were used in each of the two 806,400 cubic foot tobacco warehouses. One warehouse had a permanent piping system, the other had not. All gas samples drawn from 2-foot and 6-foot levels were taken from confined areas between and under "nested" hogsheads of tobacco. Samples drawn from 12-foot elevations were above the top of the upper tier of hogsheads. Sample number 13 was drawn from an 18-foot level, just under the roof of the warehouse, within 6 inches of the ceiling.

From one end of the non-piped warehouse, the gas from eleven 75-pound cylinders (net weight) was pumped through the hose lines laid along the central aisle. The gas was released through four hose lines having a total of 32 nozzles. It required 40 minutes to release the fumigant.

In the piped warehouse, figure 1, the 11 cylinders of gas were pumped simultaneously from both ends of the building. At each end, two cylinders of gas were released through each of two permanently installed pipe lines. Each pipe line was approximately 250 feet long and was fitted with 14 nozzles. The three remaining cylinders were pumped from one end of the building through two rubber hose lines that extended the length of the aisle. Each of the lines was fitted with eight nozzles. It required approximately 30 minutes to release the HCN in the building.

As shown in table 1, the average concentration of fumigant taken from corresponding stations from the two buildings was nearly equal. However, the recoveries of fumigant from the piped warehouse for the first hour of fumigation showed a considerably higher degree of uniformity. The spread in concentration between the high and low samples was 6.6 ounces from the non-piped section, compared with 2.7 from the piped. In 4 hours the variation between the high and low concentration in the two buildings narrowed to 2.7 and 2.4 ounces, respectively. After 4 hours, the concentration of HCN was fairly uniform in both warehouses.

There was no indication of stratification in either of the two test warehouses. In the non-piped building, 6 of the 13 sampled areas were below average concentration for the building. Two were at each of the 2-, 6-, and 12-foot elevations, From the piped warehouse, four areas were below average concentration. One each of the 12-foot and

Table 1.—Ounces of HCN recovered from a piped and from an unpiped tobacco warehouse fumigated at a dosage of 16 ounces per 1000 cubic feet.

Station No.[a]	Ounces of Hydrogen Cyanide Recovered per 1000 Cubic Feet After Hours Indicated									
	Nonpiped Warehouse					Piped Warehouse				
	1	4	8	24	Total	1	4	8	24	Total
	2 feet above floor									
1	6.0	4.8	4.2	2.7	17.7	8.7	6.0	3.9	2.1	20.7
4	7.8	6.6	4.5	2.4	21.3	10.5	7.8	5.1	2.7	26.1
11	9.6	6.3	5.4	2.1	23.4	7.8	5.4	4.5	2.7	20.4
12	10.2	7.5	5.4	2.4	25.5	9.0	7.8	5.4	2.4	24.6
	6 feet above floor									
2	9.3	7.5	5.1	2.7	24.6	9.0	7.2	5.7	3.0	24.9
5	10.5	6.9	4.8	2.7	24.9	9.6	7.2	5.1	2.4	24.3
7	6.3	5.7	5.1	2.1	19.2	7.8	5.4	4.2	2.1	19.5
9	8.4	5.7	3.9	2.4	20.4	10.2	7.5	5.4	2.7	25.8
	12 feet above floor									
3	8.7	6.9	5.4	2.7	23.7	9.0	7.2	4.8	3.0	24.0
6	8.1	6.9	3.9	2.4	21.3	9.9	7.5	5.4	2.7	25.5
8	8.1	5.4	4.8	2.1	20.4	8.7	7.8	5.7	3.3	25.5
10	9.9	7.5	5.1	2.4	24.9	7.8	5.7	4.8	2.4	20.7
	18 feet above floor									
13	12.6	7.5	4.5	2.4	27.0	9.9	7.5	5.1	2.1	24.6
Average	8.9	6.6	4.8	2.4	22.6	9.0	6.9	5.0	2.6	23.6

[a] Location of samples removed from air space.

6-foot elevations, and the remaining two at the 2-foot level. Although HCN is a lighter-than-air gas, an abnormally high concentration was not found at the 18-foot level in either of the two warehouses.

In this experiment, as well as in previous tests, the lowest concentration of gas was at the end of the building most distant from where the gas was pumped. This was shown in the concentration of fumigant in the non-piped building at locations 1, 7, 8, and 9.

CONCENTRATION AND EFFECT OF FUMIGANT.—The data in figure 2 present the average gas concentrations collected from 10 sampling areas from each of two tobacco warehouses. The figure presents a vivid picture of the advantage of an additional 16-ounce booster shot over the normal 16-ounce dosage. In the warehouse treated with the extra 16 ounces of HCN, nearly as high, or a higher concentration of gas was maintained for 24 hours, as was found at the end of the first hour in the warehouse treated with the usual dosage. After 24 hours the difference in concentration of gas between the two houses was still nearly 4 ounces, and it was 72 hours before the gas concentration in the two buildings became approximately equal. However, it should be mentioned that in past experiments, when using a 16-ounce dosage, the concentration of gas found at the end of the first hour usually ranged between 8 and 10 ounces. The 6-ounce reading recovered at the end of the first hour of fumigation in this test, therefore, was lower than usual.

The benefits of the booster shot were reflected in the mortality of the cigarette beetle larvae exposed in 10 test spikes in each warehouse. The data from table 2 indicate that both treatments were effective up to a depth of 3 inches in the tobacco. Beyond 3 inches, the booster shot showed an appreciably higher rate of mortality. At a depth of 7 inches, the heavier dosage gave an average mortality of 61% as compared with 21% from the other warehouse.

The relative insect infestation, as shown by weekly trap records presented in table 3, indicated a definite improvement in the warehouse where the booster shot was used. One week previous to fumigation, 264,000 beetles

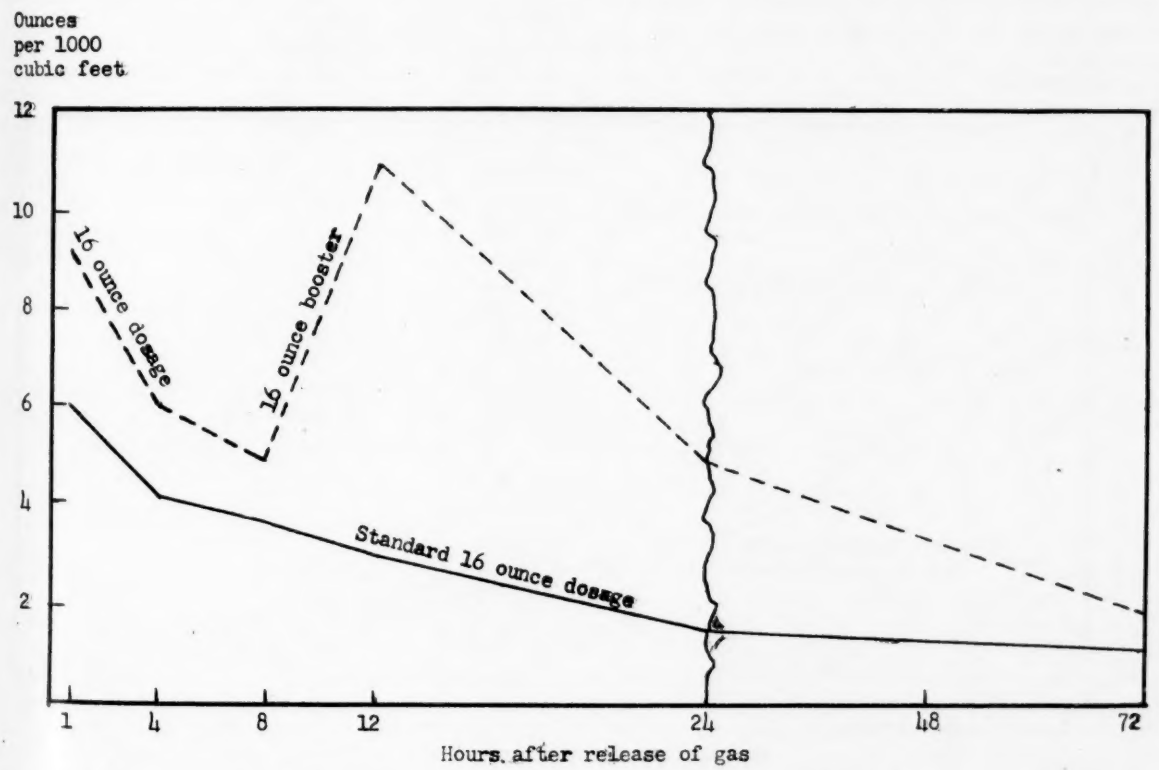


FIG. 2.—Relative concentrations of HCN in two tobacco warehouses. One was fumigated with the standard 16 oz./1000 cu. ft. dosage, and one was fumigated with an initial 16 oz./1000 cu. ft. dosage followed by a 16-ounce booster shot at the end of 8 hours.

Table 2.—Mortality[a] of 1000 cigarette beetle larvae which were exposed at five depths in 10 hogsheads of tobacco fumigated with HCN at a dosage of either 16 or 32 ounces per 1000 cubic feet.

Ounces of HCN	Per Cent Mortality at Depth Indicated (Inches)				
	1	3	5	7	9
16	100	91	52	21	13
16 initial plus 16 booster after 8 hours	100	100	86	61	34

[a] Mortality of 250 control larvae was 2%.

Table 3.—Weekly trap catches of cigarette beetles in warehouses of flue-cured tobacco fumigated with HCN.

Date of Count		Dosage—(Ounces per 1000 Cu. Ft.) 16 Initial Plus 16 Booster		16
		Before fumigation		
June 5		264,000		38,000
		After fumigation		
June 19		600		400
26		218		340
July 3		8,400		900
10		900		3,400
17		600		8,400
24		1,200		Second fumigation
31		900		5
Aug. 7		600		700
14		240		320
21		72		164
28		480		370
Sept. 4		2,400	*M*	4,200
Total	(12 weeks)	16,610	(11 weeks)	19,199

were trapped in the section that received the booster shot using a total dosage of 32 ounces of fumigant. In the section treated with the standard 16-ounce dosage, 38,000 were trapped. During the first 3 weeks following fumigation, more beetles were trapped in the building that received the heavier dosage of fumigant. However, after that time the trend reversed, and the catch from each warehouse the fifth week following fumigation was 600 beetles from the warehouse receiving the heavier dosage compared with 8400 from the warehouse receiving the regular, lighter dosage.

After 6 weeks it was necessary to re-fumigate the warehouse treated with the 16-ounce, standard dosage. Twelve weeks after the first fumigation, the warehouse that was given the booster shot had a weekly trap catch of 2400 beetles. The weekly catch for the same period from the warehouse that received two fumigations, each of 16 ounces, was 4200 beetles. Better control was obtained with one fumigation, using a total of 32 ounces, than from two fumigations, each of 16 ounces per 1000 cubic feet.

A similar experiment was conducted at a later date with three test warehouses. In one warehouse a 24-ounce dosage of HCN was released; in a second warehouse a 32-ounce dosage was used; and in the third an initial dosage of 16 ounces was released, followed in 8 hours with a 16-ounce booster shot. The data in figure 3 illustrate the average concentration from six sampling points for each

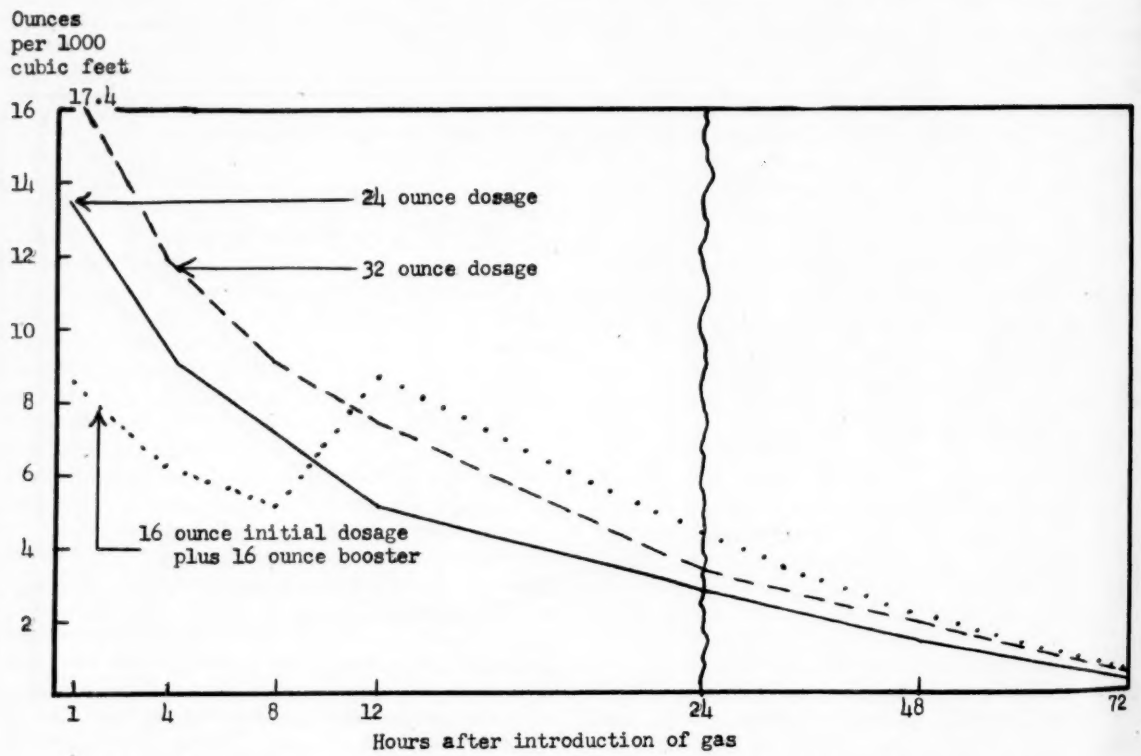


FIG. 3.—Relative concentrations of HCN in three tobacco warehouses. One was fumigated with 24 oz./1000 cu. ft.; one with 32 oz./1000 cu. ft., and one with a 16 oz./1000 cu. ft. followed by a 16-ounce booster shot at the end of 8 hours.

Table 4.—Mortality[a] of 1000 cigarette beetle larvae which were exposed at five depths in 8 hogsheads of tobacco fumigated with HCN at a dosage of either 24 or 32 ounces per 1000 cubic feet.

Ounces of HCN	Per Cent Mortality at Inches Indicated				
	1	3	5	7	9
24	100	84	44	28	19
32	100	100	88	53	31
16 initial plus 16 booster after 8 hours	100	84	64	51	37

[a] Mortality of 250 control larvae was 2%.

warehouse. As shown in this figure, the curves for the 24-ounce and 32-ounce dosages tended to parallel each other throughout the 72-hour fumigation. With the exception of the reading at the end of the first hour, there was a differential between the two warehouses of 2 ounces or less for the entire 72-hour fumigation period. The warehouse that received the initial 16-ounce dosage for the first 8 hours showed a typical relatively low concentration of HCN. However, following the booster shot the concentration of gas for the remainder of the fumigation was higher in that warehouse than in the other two test sections. If concentration X time values are computed for the time intervals at which samples were taken, little difference in value is shown between the two 32-ounce dosages, one of which was introduced as a 16-ounce initial dosage followed by a 16-ounce booster shot.

In the three warehouses, at a depth of 3 inches, mortality of the cigarette beetle larvae exposed in test spikes ranged from 83 to 100%. At depths of 5 inches, or more, appreciably higher mortality was maintained in the two warehouses treated with the heavier dosages (see table 4.) Data from the test spikes indicate that possibly slightly better penetration of HCN was obtained by releasing all 32 ounces of HCN at one time. However, control of the natural insect population of cigarette beetles, as reflected by the catches from the suction-light traps indicated that both the 32-ounce applications of HCN were reasonably effective. Data in table 5 show that for the first 6 weeks following fumigation, nearly three times as many beetles were trapped in the warehouse that was treated with 24 ounces as in either of the buildings that received the 32-ounce dosage. (Total trap catches were 7111 beetles for the 24-ounce dosage, 2140 for the 32-ounce dosage introduced at one time, and 1520 for the 32-ounce dosage introduced half as a booster shot.)

Table 5.—Weekly trap catches of cigarette beetles in warehouses of flue-cured tobacco fumigated with HCN.

Date of Count	Dosage—(Ounces per 1000 Cubic Feet)		
	24	32	16 Initial Plus 16 Booster
		Before fumigation	
July 17	229,000	206,000	155,000
		After fumigation	
July 31	14	6	18
Aug. 7	97	89	64
14	900	63	84
21	1,800	158	320
28	900	224	134
Sept. 4	3,400	1,600	900
Total (6 weeks)	7,111	2,140	1,520

Discussion.—After 4 hours, distribution of HCN throughout fumigated warehouses, piped or unpiped, was relatively uniform. However, some improvement in distribution probably could be obtained by increasing the number of nozzles towards the end of the fumigation lines. That would offset the higher flow-rate from the nozzles located nearest the pumping station.

A permanently installed piping system offered the advantage of a more uniform distribution of fumigant during the first 4 hours of treatment. It was during these hours that the gas concentration was at its peak, which is probably one of the primary factors influencing the depth of penetration of fumigant into the tobacco. Also, data from previous experiments indicate that through the use of a piping system, the fumigant was dispersed more rapidly, which tends to increase the effectiveness of the fumigation.

The data from these experiments indicate that a 16-ounce dosage is not sufficient to control a heavy cigarette beetle infestation in a warehouse filled to capacity with hogsheads of tobacco. Additional data are required to determine the most economical and satisfactory dosage to use under modern conditions of storage. Moreover, insect control is not the sole consideration. Higher dosages increase the hazard to the fumigators, to the warehouse employees, and to the surrounding areas in which dwellings sometimes are located.

Another factor affecting fumigation, which is worthy of further study, is the sorption of gas by the tobacco. Sorption almost certainly varies with different types and grades of tobacco, with tobacco of different moisture content, and different temperatures.

Investigations with New Insecticides for Codling Moth Control[1]

HAROLD F. MADSEN and STANLEY C. HOYT, *University of California, Berkeley*

ABSTRACT

Orchard trials with new insecticides for control of codling moth (*Carpocapsa pomonella* (L.)) on pears were conducted in two orchards in northern California during 1956. Diazinon alone, a combination of Diazinon and DDT, Thimet, Dipterex and a spore preparation of *Bacillus thuringiensis* Berliner failed to give satisfactory control. Trithion gave variable results, showing promise in one trial, but providing poor control in another. Hercules AC-528 showed some promise, and ryania, Sevin (N-methyl-1-naphthyl carbamate) and Guthion gave codling moth control comparable to the standard dosage of DDT.

During 1957, orchard trials were continued on pears, and Sevin and Guthion gave good codling moth control. Guthion also controlled the European red mite (*Panonychus ulmi* (Koch)), and both Guthion and Sevin reduced a lepidopterous leaf miner infestation to a low level. Ryania, at a low dosage, plus a spreader-sticker, failed to provide adequate reduction of fruit injury by the codling moth.

In a Newtown Pippin apple orchard, a heavy codling moth infestation was found following grower application of a full DDT program. Field trials during 1957 in this orchard with DDT strongly indicated a resistance to this compound. In the same orchard, Trithion, ryania, Sevin and ethion provided satisfactory control of a heavy infestation. Trithion and ethion also provided control of European red mite, and Sevin prevented a build-up of the apple aphid *Aphis pomi* DeG. No serious phytotoxic effects were noted on pears or apples following the use of the experimental insecticides.

For the past several seasons, orchard trials have been conducted with new spray chemicals for control of codling moth on pears and apples in northern California. The results of the earlier tests have been reported by Madsen (1956). Since that time, increased numbers of reports by growers of difficulty in controlling codling moth have been received, and in addition, there has been much interest in compounds that have an effect on the other fruit pests which are involved in the codling moth spray schedule.

Some of the complexities involved in the spray program on apples have been discussed by Cutright (1955), and there have been additional reports of codling moth resistance to DDT by Hamilton (1956). An attempt to use ryania as a selective insecticide has been reported by Clancy & McAlister (1956) who showed that the use of this material could prevent spider mite build-up. The report of resistance to TDE in the red-banded leaf roller (*Argyrotaenia velutinana* (Wlk.)) by Glass (1957) adds another factor to the overall problem.

ORCHARD TRIALS IN 1956.—One series of experimental plots was established in a Bartlett pear orchard near Courtland, California. This orchard had a history of severe codling moth in one area about 3 acres in extent. This situation was related to the fact that the codling moth emerged some 2 to 3 weeks later in this locality than in other areas. As a result, improper timing of sprays resulted in heavy infestation in previous seasons, and made this area a good one for testing new materials. In 1956, Diazinon and Trithion were tested. Wettable 25% Diazinon was used at a 2-pound dosage alone, and at 1 pound plus 1 pound of wettable 50% DDT per 100 gallons of water. This latter combination was compared with wettable 25% parathion at 1 pound plus DDT at 1 pound. The applications were made with blower sprayer equipment, and were timed by the use of bait pans. At each application, the applied gallonage averaged 710 gallons per acre. Each plot was one-half acre in size and replicated twice.

At harvest, fruit was picked from the center 16 trees in each plot and examined for worms and stings. Prior to the first picking, the dropped fruit was collected from each plot and included with the picked fruit examined to provide a figure representing the total crop infestation. The materials used, dosages, and harvest counts are summarized in table 1.

Trithion and Diazinon, as well as the Diazinon-DDT combination, failed to give commercial control in this test. The DDT-parathion combination, however, reduced the fruit damage to a reasonable figure. The 1-pound dosage of DDT alone is not sufficient to control the codling moth, as has been observed in grower-sprayed orchards, therefore it seems that Diazinon added little to the combination, whereas parathion partially overcame the poor control obtained with the low DDT dosage.

With regard to other pests, it was necessary to add an acaricide to all of the treatments for control of European red mite with the exception of Trithion which controlled this species for the season, when three treatments were applied for codling moth.

A second series of plots was established in a Hardy pear orchard near San Jose, California. This orchard had been abandoned for several seasons and thus had a high codling moth population. The rather poor condition of the trees resulted in a light crop, which somewhat intensified the infestation. It was however, an ideal spot to test new materials which had little data available with regard to harzards of spray residues, since the fruit was not marketed.

Each plot consisted of four trees with three replicates, and materials were applied with conventional ground equipment and orchard guns. The sprays were again timed by the use of bait pans. The amount of spray used per acre averaged 750 gallons.

At harvest, an attempt was made to examine 200 fruit per replicate for entries and stings, but it was not possible to attain this figure on all plots, because of the poor set of

[1] Accepted for publication January 2, 1958.

Table 1.—Results of treatments against codling moth on Bartlett pears at Courtland, Calif. 1956.

WETTABLE POWDERS[a]	POUNDS PER ACRE	NO. OF FRUIT EXAMINED[b]	ENTRIES	STINGS	PER CENT INFESTED FRUIT
Diazinon, 25%	14.2	2215	140	20	7.2
Diazinon, 25%+DDT, 50%	7.1 7.1 7.1	2326	149	28	7.6
Parathion, 25%+DDT, 50%	7.1	2140	47	6	2.4
Trithion, 25%	14.2	2304	144	20	7.1

[a] Materials applied May 1, June 1, and June 29.
[b] Total of two pickings, the first on July 24, the second on August 7 (preharvest drops included).

fruit. The materials used, dosages, and harvest counts are summarized in table 2.

Several materials, including Diazinon, Thimet, Dipterex and the spore preparation of *Bacillus thuringiensis* failed to give adequate codling moth control. Two other materials, Trithion and Hercules AC-528 gave only fair control. It is interesting to note than the combination of DDT and Diazinon at 1 pound of each material was no better than the 1 pound of DDT alone, which parallels the results obtained in the Courtland plots. Ryania and DDT both gave good control and two new materials, Sevin and Guthion, gave excellent results. Other pests, including aphids and spider mites, were present in this orchard, but populations were too low to evaluate the effect of the codling moth sprays on these pests.

ORCHARD TRIALS IN 1957.—In the 1957 season, comparisons of materials for codling moth control were continued on the same orchard in Courtland, California which was used during 1956. Two of the newer materials, Guthion and Sevin, were compared with the standard material, DDT. In addition, a reduced dosage of ryania plus a special sticker-spreader was tried in an attempt to reduce the cost of a full dosage.

At the end of the 1956 season a pear orchard near the Courtland plot was reported to have a codling moth problem in a 5-acre section and the same spray schedule was therefore repeated in this orchard.

The applications of the sprays in both orchards were timed by means of bait pans, and it was found that a late emergence of first brood moths also occurred in the second orchard. In 1957, this emergence was 2 weeks later than that reported from other areas.

In the first orchard (the Amaro ranch), plots were one-half acre in size with two replications, and the materials were applied with blower-sprayer equipment. At each application, the gallonage per acre averaged 750. Records were taken in the same manner as for the data presented in table 1 and the materials used, dosages, and harvest counts are summarized in table 3.

Guthion gave excellent control of codling moth in this orchard, reducing the infestation to a very low figure. Sevin was not so effective as Guthion in this test, although it provided commercial control as did the standard dosage of DDT. Ryania, at the reduced dosage, did not provide adequate control even though a special spreader-sticker was used.

European red mite required treatment in this orchard in late June, and it was necessary to apply an acaricide to all plots with the exception of those treated with Guthion. This material controlled the European red mite

Table 2.—Results of treatments against codling moth on Hardy pears. San Jose, Calif. 1956.

MATERIALS	DOSAGE PER 100 GALLONS[a]	NO. OF FRUIT COUNTED	ENTRIES	STINGS	PER CENT INFESTED FRUIT
DDT, 50%	2	600	2	4	1.0
DDT, 50%	1	600	15	8	3.6
Diazinon, 25%+DDT, 50%	1	564	14	7	3.7
Diazinon, 25%	2	600	26	8	5.6
Thimet	1[b]	600	28	33	10.1
Hercules AC-528, 25%	2	600	8	6	2.3
Sevin, 50%	1.5	553	2	1	0.6
Trithion, 25%	2	600	20	6	4.3
Dipterex, 50%	4	516	32	19	9.8
Ryania, 100%	6	586	7	4	1.8
Guthion, 25%	2	600	2	1	0.5
Bacillus thuringiensis	100[c]	600	80	18	16.3
Check No. 1	—	600	92	32	20.5
Check No. 2	—	600	115	21	22.6

[a] Applied on May 1, May 28, and July 9. Pounds of wettable powder except as noted.
[b] One quart of 48% emulsion concentrate.
[c] Grams of spore preparation.

for the season, when applied three times for codling moth control.

In the second orchard (Van Loben Sels) the plots used were each 1 acre in size and were not replicated. Materials were applied with blower-sprayer equipment, and the applied gallonage per acre averaged 700.

The same timing was used as for the first orchard with one exception. The grower applied the first DDT spray 2 weeks earlier than the first application of test materials, since this was his standard procedure. At that time, the bait pans had shown no moths, although a first flight had occurred in other areas. Subsequent DDT treatments were applied at the same time as the experimental compounds.

The fruit samples at harvest were taken in the same manner as previously described, and the materials used, dosages, and harvest counts are summarized in table 3.

The codling moth population was lower in this orchard, and as a result, all materials gave commercial control. Guthion and Sevin, however, were better than the reduced dosage of ryania and the standard DDT treatment. The early timing of the first DDT spray may have been responsible for the results obtained with this material, and also may explain why a codling moth problem existed in this orchard. These two orchards, within one-half mile of each other, showed the same late emergence of first brood codling moth which complicated the control program by making necessary a change in the standard timing of treatments.

In the Van Loben Sels orchard, a fairly heavy population of a tentiform leaf miner, *Lithocolletis* sp., was present, and the treated plots were checked for leaf damage in late June. They were compared with DDT, as previous work had shown that it does not give control of the miner. The leaf counts showed that very few mines were present on both the Guthion- and Sevin-sprayed trees. Ryania, however, did not provide control and the leaves sprayed with it were as heavily infested as those on the DDT plots.

In the fall of 1956, a report was received of an apple orchard near Soquel, California, which had suffered heavily from codling moth damage. Investigation at harvest disclosed over 60% infested fruit following a DDT program of 5 applications spaced 3 weeks apart

Table 3.—Results of treatments against codling moth on Bartlett pears in Amaro and Van Loben Sels orchards. Courtland, Calif. 1957.

Pounds of Wettable Powder per Acre	Harvest Fruit Counts			
	Fruit Examined	Codling Moth		Per Cent Infested Fruit
		Entries	Stings	
Amaro Orchard[a]				
Guthion, 15%, 22.7	1133	2	1	0.1
Sevin, 50%, 10.6	1278	31	7	2.4
Ryania, 100%, 21.3+ spreader-sticker, 1.7	1197	111	24	9.2
DDT, 50%, 14.2	1093	14	2	1.2
Van Loben Sels Orchard[b]				
Guthion, 15%, 22	1119	0	0	0.0
Sevin, 50%, 10.5	1086	2	6	0.2
Ryania, 100%, 20.7+ R.S. spreader-sticker, 1.7	1207	22	4	1.8
DDT,[c] 50%, 14	1087	24	8	2.2

[a] Materials applied May 14, June 6, and June 26 and the results are based on pickings made July 12 and July 27 (preharvest drops included).
[b] Materials applied May 13, June 6, and June 26 and the results are based on pickings made July 12 and July 31.
[c] First application made May 1.

As the situation suggested a resistance problem, overwintering larvae were collected for laboratory testing, and a series of experimental plots were set up in the most heavily infested section of the orchard in the spring of 1957. The apple variety involved was principally yellow Newtown Pippin, with a few scattered Red Delicious trees present. Each plot consisted of four trees, with three replications, and materials were applied with conventional ground equipment and orchard guns. At each application, the applied gallonage per acre averaged 700.

The sprays were timed by means of bait pans, and it was necessary to apply four treatments during the season. At harvest, 500 fruits from each replicate, a total of 1500 fruits per treatment, were examined for entries and stings. The materials used, dosages, and harvest counts are summarized in table 4. The harvest counts demonstrated that the DDT-sprayed plots had over 30% entries, strongly indicating a DDT resistance. When parathion was included with the DDT, the numbers of entries were reduced considerably. Ryania, Trithion, and ethion all gave commercial control in a heavy infestation. The results with Trithion were better than those previously obtained, and may be related to the use of a flowable formulation rather than a wettable powder. Sevin was outstanding in this trial, and very few infested fruits were in evidence. It will be of interest to note that analyses for insecticide residues on the apples from the above plots showed an initial deposit of over 5 p.p.m.

Table 4.—Results of treatments against codling moth on Newtown pippin apples, Manildi Orchard. Soquel, Calif. 1957.

Materials	Dosage[a] per Acre	Harvest Fruit Counts, Oct. 14			
		Fruit Counted	Codling Moth		Per Cent Infested Fruit
			Entries	Stings	
DDT, 50%	13.6	1500	470	33	31.3
DDT, 50%+ Parathion, 25%	13.6 6.5	1500	98	23	6.5
Ryania, 50%	40.2	1500	52	51	3.4
Sevin, 50%	11.4	1500	4	2	0.2
Trithion	6.8	1500	33	13	2.2
Ethion, 25%	15.4	1500	18	12	1.2

[a] Sprays applied May 6, June 10, July 18, and Sept. 12. Pounds of wettable powder except the dosage of Trithion is given in pints of 50% emulsion concentrate.

of Sevin. The analyses of samples from the pear plots previously discussed showed only a little over 2 p.p.m. Sevin as an initial deposit. The difference in the control obtained on the pears and apples may be related to problems of deposit on the different fruits.

The apple aphid and the European red mite were also present in this orchard, and there was an opportunity to evaluate the materials applied against the codling moth on these pests. Sevin provided good control of the apple aphid, as did the DDT-parathion combination. Ethion and Trithion, although phosphates, gave only partial control, and ryania and DDT were ineffective.

The European red mite became abundant enough to require treatment in early July on all plots except those treated with ethion and Trithion. These two materials reduced populations to a very low level and maintained control throughout the season.

No serious phytotoxic effects were noted on either pears or apples with the materials used during 1957. Trithion caused a few dark spots on one side of yellow Newtown Pippen apples, but the injury was very shallow and not present on all fruits.

References Cited

Clancy, D. W., and H. J. McAllister. 1956. Selective pesticides as aids to biological control of apple pests. Jour. Econ. Ent. **49**(2): 196–202.

Cutright, C. R. 1955. Increasing complexities in the apple spray program. Jour. Econ. Ent. **48**(3): 304–6.

Glass, E. H. 1957. The occurrence of resistance to TDE (DDD) in the red banded leafroller. Jour. Econ. Ent. **50**(5): 674–6.

Hamilton, D. W. 1956. Resistance of the codling moth to DDT sprays. Jour. Econ. Ent. **49**(6): 866–7.

Madsen, Harold F. 1956. Codling moth control on pears 1954–55. Jour. Econ. Ent. **49**(4): 467–70.

Laboratory Screening Tests of Animal Systemic Insecticides[1]

R. O. Drummond, *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*

Abstract

Systemic, insecticidal properties of candidate insecticides received at the Kerrville, Texas, laboratory are determined by the following short-term laboratory test. Guinea pigs, infested with nymphal lone star ticks (*Amblyomma americanum* (L.)) and screw-worms (*Callitroga hominivorax* (Cqrl.)) are treated orally or subcutaneously with the compounds. After treatment, stable flies (*Stomoxys calcitrans* (L.)) are fed on the guinea pigs.

From many compounds which have been screened, results of tests with nine are presented to illustrate variations in toxicity to the arthropods and lethality to the guinea pigs. Am. Cyanamid 12,880 (*O,O*-dimethyl *S*-(N-methylcarbamoylmethyl) phosphorodithioate), Bayer 23,129 (*S*-2-(ethylthio)ethyl *O,O*-dimethyl ester phosphorodithioic acid), and Bayer 23,453 (*S*-2-(ethylsulfinyl) ethyl *O,O*-dimethyl ester phosphorodithioic acid) were toxic to all three species. Bayer 21/199 and Dow ET-57 were toxic to screw-worms and stable flies but not lone star ticks. Hercules AC-528, Tedion, Thiodan, and Trithion were ineffective.

Compounds which showed promise in tests with guinea pigs were tested further in sheep and goats, being administered as drenches to sheep infested with screw-worms and to goats infested with screw-worms and lone star ticks. After treatment, stable flies were fed on the goats. In addition to entomological data, this test is used by cooperating veterinarians to obtain toxicological data.

Am. Cyanamid 12,880, Bayer 23,129, and Dow ET-57, used in tests with sheep and goats, were all effective against screw-worms, and 12,880 was also effective against the other arthropods.

In the last 2 years there has been rapid progress in the development of systemic insecticides for the control of cattle grubs. Dow ET-57 as a drench gave 97% control of both *Hypoderma lineatum* (De Vill.) and *H. bovis* (L.) in a test at Kerrville, Texas, as reported by McGregor & Bushland (1957). Jones *et al.* (1957) and Roth & Eddy (1957) have also obtained encouraging results with this material. Bayer 21/199, when administered as a spray, gave complete control of grubs in a small-scale field test (Brundrett *et al.* 1957). Both these materials affected cattle grubs while they were still inside the animals' bodies and prevented grubs from encysting in their backs and damaging meat and hides. They are considered to be systemic insecticides because, although administered differently, they were transported throughout the animals' bodies and were toxic to grubs within them.

Control of cattle grubs is one of the goals of research with systemic insecticides. However, the year-long life cycle and seasonal occurrence of cattle grubs, plus the costs of buying and maintaining cattle, make studies with cattle time-consuming and expensive. Most research to find systemic insecticides has been done with other host-parasite relationships. The first report of control of insects with a systemic insecticide was by Lindquist *et al.* (1944), in which rabbits were made toxic to bedbugs (*Cimex lectularius* L.) by treatment with DDT or pyrethrum. This work was followed by reports (de Meillen 1946, Garnham 1947, Knipling *et al.* 1948) that rabbits, mice, and other animals were made toxic to lice, mosquitoes, bedbugs, and ticks by administration of gamma isomer of BHC and some of the indandione compounds.

In 1946 a screening program using short-term, inexpensive tests with laboratory animals was established at the Kerrville laboratory. At first this program consisted of *in vivo* and *in vitro* tests against first-instar cattle grubs (Barrett & Wells 1948). Hundreds of compounds were screened by this procedure, but none was found that seemed worthy of testing on cattle.

A report by de Toledo & Saur (1950) that BHC was systemically active against the human bot fly (*Dermatobia hominis* (L., Jr.)) in cattle stimulated research on a number of chlorinated hydrocarbon insecticides as systemics. Lindquist *et al.* (1953) reported that aldrin, dieldrin, and lindane differed in effectiveness against bloodsucking insects feeding on treated white mice and livestock. McGregor *et al.* (1955) found that these compounds killed screw-worms when injected into guinea pigs and second- and third-instar cattle grubs in the backs of cattle but did not prevent encystment of new grubs.

Most of the known chlorinated hydrocarbons were tested for systemic properties at Kerrville. The *in vitro* and *in vivo* procedures were replaced with a screening test in which compounds were administered subcutaneously to guinea pigs infested with larvae of the screw-worm (*Callitroga hominivorax* (Cqrl.)). Many phosphorus compounds were also tested. Dipterex, Diazinon, Chlorthion, and Bayer 21/199 showed promise in tests with guinea pigs, but none of them when given orally to cattle prevented new grubs from encysting in their backs (McGregor *et al.* 1954).

Oral as well as subcutaneous administration was included in the screening test with guinea pigs. Also, techniques were developed for feeding stable flies (*Stomoxys calcitrans* (L.)) and adult lone star ticks (*Amblyomma americanum* (L.)) on guinea pigs. Sheep were used occasionally to test compounds which had shown promise in the test with guinea pigs. The screening program through 1955 has been reviewed by McGregor & Bushland (1956).

Since then several changes have been made in the guinea pig test and goats have been used in conjunction with sheep. This paper presents schedules of events with guinea pigs, sheep, and goats, and results of several compounds used in these tests.

Tests with Guinea Pigs.—This test requires approximately 5 days. Guinea pigs were weighed, and the hair on their bellies was removed with clippers. Small plastic containers were attached to their bodies, and 10

[1] Accepted for publication January 2, 1958.

Table 1.—Systemic activity of nine compounds used in tests with guinea pigs.

Compound	Administration	Lowest Dosage (Mg./Kg.) Lethal to Host	Lowest Dosage (Mg./Kg.) Causing Complete Kill of—		
			Screw-Worms	Stable Flies	Lone Star Ticks
Am. Cyanamid 12,880	Oral	—[a]	10	10	25
	Subcutaneous	—	5	25	50
Bayer 21/199	Oral	25	25	25	*[b]
	Subcutaneous	100	75	50	*
Bayer 23,129	Oral	—	5	10	10
	Subcutaneous	50	*	*	*
Bayer 23,453	Oral	—	10	25	25
	Subcutaneous	75	50	50	50
Dow ET-57	Oral	—	50	50	*
	Subcutaneous	—	50	50	*
Hercules AC-528	Oral	10	*	*	*
	Subcutaneous	—	*	*	*
Tedion	Oral	—	*	*	*
	Subcutaneous	—	*	*	*
Thiodan	Oral	10	*	*	*
	Subcutaneous	25	*	*	*
Trithion	Oral	50	*	*	*
	Subcutaneous	100	*	*	*

[a] Not lethal at highest dosage tested.
[b] Asterisk indicates that there was no kill at highest nonlethal dosage tested.

starved nymphal lone star ticks were placed in each. Twenty-four hours later the guinea pigs were wounded on their hips, and each pig was infested with approximately 50 newly hatched screw-worm larvae.

When the larvae had been in the wounds for 24 hours, the pigs were treated with the test insecticide, usually a 5% solution in Tween-20 (polyoxyethylene sorbitan monolaurate). A dosage of 100 mg./kg. was administered to two pigs, orally to one and subcutaneously to the other, and after 4 and 24 hours stable flies were allowed to feed on the pigs. The flies were then held for 24 hours to see if they were affected by the blood they ingested. If the screw-worms were killed by the insecticide, the guinea pigs were reinfested with newly hatched larvae at 24-hour intervals to determine the duration of residual effects.

Nymphal ticks finished engorging about 3 days after the pigs were treated. Those ticks that engorged were held until they died or molted, and those that became adults were counted as survivors. If the insecticide killed any of the arthropods or was lethal to the guinea pig at the initial dosage, the test was repeated with successively lower dosages until there was no insecticidal activity and/or no lethality to the guinea pig.

During the last year many compounds have been screened in this test. Some have been lethal to the guinea pigs, and others have shown different degrees of toxicity to the arthropods. Many compounds have been tolerated by the guinea pigs, but have also been ineffective as systemic insecticides. The effectiveness of nine compounds against the different species and their lethality to guinea pigs are summarized in table 1.

Am. Cyanamid 12,880 (*O,O*-dimethyl S-(N-methylcarbamoylmethyl) phosphorodithioate), Bayer 23,129 (S-2-(ethylthio)ethyl *O,O*-dimethyl phopshorodithioate), and Bayer 23,453 (S-2-(ethylsulfinyl)ethyl *O,O*-dimethyl phosphorodithioate) gave 100% kill of all three arthropods. Bayer 21/199 and Dow ET-57 caused mortality of screw-worms and stable flies but not lone star ticks. Hercules AC-528, Tedion, Thiodan, and Trithion had no effect on the different species.

TESTS WITH SHEEP AND GOATS.—In order to obtain more data about a compound which has shown promise in the guinea pig test, it was used as a drench in a larger animal. Sheep have been used, but presently goats have replaced sheep because goats were more resistant to bacterial infections which accompany screw-worm wounds and all three species used in the guinea pig test parasitized goats more readily than sheep. The animals were watched closely by entomologists to see if the compound was as effective as in the guinea pig test, and by veterinarians of the U. S. Department of Agriculture Animal Disease and Parasite Research Division for signs of insecticide poisoning.

The tests with sheep and goats required approximately 14 days. Four days before the animals were drenched, they were wounded and each animal was infested with 200 to 300 newly hatched screw-worm larvae. Starved adult lone star ticks were placed inside plastic containers which were attached to the goats' ears. One day before the drenching, the animals were wounded a second time and newly hatched screw-worm larvae were placed in these wounds, and starved nymphal lone star ticks were placed in the containers on the goats' ears. The animals were weighed and drenched with aqueous solutions of the insecticide. If the compound was not soluble in water, a wettable powder or emulsion concentrate was formulated, and the resulting mixture was dispersed in water for the drench. The initial dosage was 100 mg./kg. unless a compound exhibited a high mammalian toxicity in the tests with guinea pigs. After 4 and 24 hours starved stable flies were allowed to feed on the goats, and then held for 24 hours to see if they were affected by the blood they ingested. If the screw-worm larvae were killed by the insecticide, wounds were reinfested with newly hatched larvae every 24 hours until larvae lived normally in the wounds. From 2 to 10 days after the drenching, nymphal and adult ticks finished engorging and detached from the goats. The nymphs were held to see if they molted normally, but the adults that engorged were not held for further observations.

Three of the compounds that showed promise in the guinea pig test were used in tests with sheep and goats. Am. Cyanamid 12,880 was not lethal to sheep or goats, but killed both nymphal and adult ticks at dosages as low as 50 mg./kg. and stable flies and screw-worms as low as 25 mg./kg. Bayer 23,129 was lethal to goats and sheep as low as 25 mg./kg., but killed screw-worms at 10 mg./kg. and was not toxic to stable flies and lone star ticks. Dow ET-57 was not lethal to the goats and sheep, nor did it kill lone star ticks or stable flies, but it caused 100% mortality of screw-worms at 100 mg./kg.

REFERENCES CITED

Barrett, W. L., and R. W. Wells, 1948. Transplantation of *Hypoderma* larvae and testing chemicals for control of larvae in experimental hosts, Jour. Econ. Ent. 41(5): 779–82.

Brundrett, H. M., W. S. McGregor, and R. C. Bushland. 1957. Systemic cattle grub control with Bayer 21/199 sprays. Agric. Chemicals 12(6): 36–37, 123, 125.

Garnham, P. C. C. 1947. Mortality of *Aedes aegypti* feeding on rabbits receiving oral "Gammexane." Nature 160(4057): 156–7.

Jones, R. H., H. M. Brundrett, and R. D. Radeleff. 1957. Ranch tests against cattle grubs with the systemic

insecticide Dow ET-57. Agric. Chemicals 12(7): 45, 95–6.

Knipling, E. F., R. C. Bushland, F. H. Babers, G. H. Culpepper, and E. S. Raun. 1948. Evaluation of selected insecticides and drugs as chemotherapeutic agents against external blood-sucking parasites. Jour. Parasitol. **34**(1): 55–70.

Lindquist, A. W., E. F. Knipling, H. A. Jones, and A. H. Madden. 1944. Mortality of bed bugs on rabbits given oral dosages of DDT and pyrethrum. Jour. Econ. Ent. **37**(1): 128.

Lindquist, A. W., A. R. Roth, R. A. Hoffman, and W. W. Yates. 1953. Chemotherapeutic use of insecticides for control of bloodsucking insects. Jour. Econ. Ent. **46**(4): 610–4.

McGregor, W. S., R. D. Radeleff, and R. C. Bushland. 1954. Some phosphorus compounds as systemic insecticides against cattle grubs. Jour. Econ. Ent. **47**(3): 465–7.

McGregor, W. S., R. D. Radeleff, R. C. Bushland, and H. V. Claborn. 1955. Dieldrin, aldrin, and lindane as systemic insecticides against cattle grubs and screwworms. Agric. Chemicals **10**(1): 34–6, 123.

McGregor, W. S., and R. C. Bushland. 1956. Research on the use of systemic insecticides for the control of livestock pests. Jour. Econ. Ent. **49**(1): 86–88.

McGregor, W. S., and R. C. Bushland. 1957. Tests with Dow ET-57 against two species of cattle grubs. Jour. Econ. Ent. **50**(3): 246–9.

deMeillon, B. 1946. Effect on some bloodsucking ectoparasites of "Gammexane" when fed to a rabbit. Nature **158**(4023): 839.

Roth, A. R., and G. W. Eddy. 1957. Tests with Dow ET-57 against cattle grubs. Jour. Econ. Ent. **50**(3): 244–6.

deToledo, A. A., and H. F. G. Saur. 1950. The effects of certain chlorinated insecticides on the warble fly. Biologico **16**(2): 25–34.

Induced Increase of Soft Scales on Walnut[1]

A. E. MICHELBACHER and STEPHEN HITCHCOCK, *University of California, Berkeley*

ABSTRACT

One of the most serious problems confronting walnut growers in California is the marked increase in the population of soft scales induced by insecticides. The trouble developed because of the interference by certain insecticides, such as DDT, with effective parasitism by hymenopterous parasites. Apparently because of their small size the parasites are adversely affected by relatively small amounts of insecticide. As a result, the problem has developed not only from treatments applied directly to walnut orchards, but is also associated with drift of insecticides, through orchards, when adjacent fields or even distant fields are treated.

Three species of soft scales are involved; frosted scale (*Lecanium pruinosum* Coq.), European fruit lecanium (*L. corni* Bouché) and the calico scale (*L. cerasorum* Ckll.). The first two occur as a complex. There appear to be typical forms and atypical forms that more or less blend into one another. The typical form of the frosted scale was more susceptible to parathion than the typical form of the European fruit lecanium.

The scales were most easily controlled during July and August at which time 2 pounds of 25% parathion wettable powder plus three-fourths gallon of light summer oil emulsion in 100 gallons of water per acre applied with an air carrier sprayer was effective against all three species. After leaf drop and until about the first of March all three species were effectively controlled with 5 pounds of 25% parathion wettable powder plus 2 to 4 gallons of oil emulsion per acre, applied with an air carrier sprayer. Control with insecticides was not possible following the second molt which usually occurs during March.

One of the most serious problems facing growers has been the marked increase in soft scales in walnut orchards during recent years. It was early recognized that applications of DDT to control the codling moth had the tendency to induce an increase in the scale population (Michelbacher *et al.* 1946, Middlekauff *et al.* 1947). The frosted scale (*Lecanium pruinosum* Coq.) appeared to be the principal species and investigations by Bartlett & Ortega (1952) and Michelbacher (1955) demonstrated that the increase in the scale population resulted from an interference of DDT with effective parasitism by hymenopterous parasites, principally *Metaphycus californicus* (Howard). A study of the problem definitely showed that a relationship existed between the dosage of DDT applied, and the rate of increase. At the lower levels the rise in the scale population was much less pronounced than it was at the higher rates. As a matter of fact it was believed that destructive outbreaks of the scale could be avoided if the dosage of DDT applied to control the codling moth did not exceed 4 pounds of actual material per acre. Continued use of DDT proved that this was not the case and additional investigations showed that other factors were involved. It it known that other insecticides besides DDT are responsible for inducing increases in the scale population. However, it should be stated that there are some insecticides which suppress the scale-population and thus tend to mask the effect of the scale-inducing agent. For example, parathion when used for summer control of the walnut aphid (*Chromaphis juglandicola* (Kltb.)) behaves in this manner.

At first there were only scattered complaints concerning increases in the scale population. A number of these were investigated and in most cases it appeared as if parasites were about to control the situation (Michelbacher 1955). This probably would have occurred if treatments responsible for the increase had been discontinued. Instead of improving, the scale problem mounted and by 1956 it reached very serious proportions. The

[1]Accepted for publication January 2, 1958.

trouble was traced to three principal sources: (1) treatments applied directly to walnut orchards for pest control; (2) treatments applied to fields adjacent to walnut orchards, and where there was a drift of insecticides through the orchard and (3) a distant dust drift that occurs in extensive agricultural areas where scattered fields of various crops are treated morning after morning during the summer (Michelbacher & Hitchcock 1957).

SPECIES OF SCALE INVOLVED.—It has long been known that two species of soft scales commonly occur on walnut in northern California. These are the frosted scale, *Lecanium pruinosum* and the calico scale *Lecanium cerasorum* Ckll. The latter for a period of time after its establishment in the San Joaquin valley threatened to become a destructive pest (Serr 1933). These two species are very characteristic and are easily separated from one another at all stages of their development.

However, the frosted scale does not occur on walnut as a pure culture. This fact came to light when attempts were made to control the pest in midFebruary of 1956 with 25% parathion wettable powder applied at the rate of 5 pounds per acre. Following application a number of surveys were conducted in order to determine the effectiveness of the treatment. Mortality counts obtained were very confusing. On some twigs practically all the scales had been killed while on others the mortality was very low. An examination of the trees indicated that the observed difference was not caused by faulty application. However, in April when the surviving scales began rapid growth it become apparent that a second species was present on the trees almost in a pure culture. Specimens were sent to Mr. Howard L. McKensie, of the California State Department of Agriculture, who identified them as the European fruit lecanium (*Lecanium corni* Bouché). The parathion treatment almost completely destroyed the frosted scale population, but apparently had no great effect upon the European fruit lecanium. The culture of the latter selected out by the parathion treatment could be easily separated from typical frosted scale only during the period of rapid growth that follows the second molt in early spring.

The investigations of 1956 showed that there was a complex of species involved. In the spring of 1957 walnut orchards throughout northern California were observed in association with Mr. McKenzie. Typical and atypical forms of both the frosted scale and the European fruit lecanium were found. The problem proved most confusing and mounted material was sent to Dr. Harold Morrison, Coccidologist, United States Department of Agriculture, who in turn was unable to solve the problem. From the observations made it appears that atypical forms of both the European fruit lecanium and the frosted scale react to the parathion treatment as the typical form of the frosted scale does. However, the study emphasized the need for correct identification, because the parathion treatment was not effective against the European fruit lecanium. From observations made it seemed possible that in a mixed culture the frosted scale was better able to survive than the European fruit lecanium. However, if the former was removed by treatment then the latter was able to expand its population rapidly.

GENERAL LIFE HISTORY.—All three species of soft scale have but a single generation a year. The eggs hatch in May and June and the young crawl out from under their mothers and settle on the new growth. On the leaves they settle on both the lower and upper surfaces. Although capable of movement they stay in one place, mostly. In the fall most individuals move back to the twig growth before the leaves drop. They cast their skins and remain principally on the undersides of the twigs throughout the winter as elongated, oval, brown individuals that measure about one-sixteenth inch in length. In March they cast their skins again after which growth becomes rapid. Eggs are produced in April and early May after which the mother scale dies. The eggs hatch and another generation is started. Development is one to several weeks more rapid in the case of calico scale and the European fruit lecanium than it is with the frosted scale.

EXPERIMENTAL METHODS.—Two methods were used in determining scale populations. During the dormant season and until the eggs hatched, the number of individuals on selected areas of twig growth were counted. The sample region consisted of the basal 2 inches of the twig growth of the past season. Although a record was made of all individuals encountered, only those that fell in the live category are considered in this discussion. In cases when effective control measures were applied, the area examined was increased to 10 inches and the number of individuals present per 2 inches was calculated by dividing the number found by 5. Increasing the length of the sample area to 10 inches was necessary because the scale population was sometimes reduced to a level where none would be taken on a series of 2-inch samples although a general search would show that a few individuals had survived the treatment. Further, post-treatment surveys were not made until spring after the scales had begun to make rapid growth. This was done in order that the few survivors could be more easily seen. The number of twigs examined per plot on each survey was 25, and in some cases the number was increased to 50. The twig samples were gathered at random from the lower branches of the trees.

After the eggs hatched and until the leaves dropped from the trees, the scale population was determined by counting the number of individuals found on impressions made on leaflets. A beveled edged tubing having an inside diameter of 15 mm. was used, and three impressions were made along the mid rib of a leaflet. Scales on both the upper and lower surfaces of the marked area were counted. For determination of populations the anteapical leaflets were selected, and they were gathered at random from the lower branches of the trees. At least 25 leaflets or 75 sample areas were examined in each plot on each survey.

DESTRUCTIVENESS.—Heavy infestations of soft scales seriously injured the trees. Vigor and size of leaf were greatly reduced. This was reflected in a lowering of the quantity and quality of the nut crop. The full impact caused by large scale populations in experimental orchards has been observed. Under most conditions it was difficult to determine accurately the real damage caused by a single sucking pest because of the presence of others. With walnuts the situation is usually complicated by the presence of the walnut aphid, spider mites, or armored scales. In the experimental areas in question the plots

Table 1.—Average number of live scales per leaf sample,[a] on October 6, 1956 in plots treated with different aphicides in the experimental orchard at Linden, Calif.

Insecticide	Number of Years Applied to Plots	Average No. of Live Scales per Sample: Frosted Scale and European Fruit Lecanium Complex	Average No. of Live Scales per Sample: Calico Scale
Schradan	1	1.04	0.04
Schradan	2	1.13	0.10
Schradan	3	0.14	0.17
Parathion	3	0.02	0.01
Malathion	3	0.10	0.03
BHC-demeton	2	1.00	0.06

[a] Leaf samples 15 mm. in diameter.

were practically free of all pests with the exception of soft scales. This was made possible through an application of schradan (OMPA) which freed the plots of the walnut aphid for the entire growing season. Such plots, where heavily infested (20 to 40 scales per leaf sample), looked unproductive when compared with surrounding plots where both scales and aphids were controlled. A survey of commercial orchards also showed that those heavily infested with scales lacked vigor and the growers complained about unsatisfactory yields. In some orchards the twig growth was caked with scales. Scale counts made during the dormant season in heavily infested orchards frequently averaged from 100 to more than 200 scales per 2-inch twig sample.

RELATION OF INSECTICIDES TO INDUCED INCREASE IN SCALE POPULATION.—One of the basic reasons for the increase of soft scales to major proportions on walnut is that their principal natural enemies are very small hymenopterous parasites which are easily disturbed by small amounts of insecticides. If left undisturbed they have the ability, through their effective action, to keep the scale population much below an economic level.

The parasites are particularly affected adversely by insecticides, especially if present when the parasites are in the adult stage. The adverse action of DDT and similar materials has been demonstrated on numerous occasions. In general it might be concluded that any materials that adversely affect the parasites, but are ineffective against the host, are likely to favor an increase in the pest's population.

A situation has developed in which the systemic insecticides demeton (Systox) and schradan have been associated with the increase in the scale populations. Both of these materials appeared to exert a strong suppressing action upon the scale population, more marked in the case of schradan than with demeton (Michelbacher & Hitchcock 1957). Despite this action, highly destructive scale populations have developed in every case where demeton has been used in experimental plots in combination with the codling moth spray to control the walnut aphid. Except in the case of the calico scale, schradan tends to hold the soft scale population in control. The beneficial action becomes more marked with increased dosage. However, little suppressive action can be expected if schradan has been applied to foliage that has hardened or where large populations are already present. The peculiar action of schradan through 1956 in tending to increase the calico scale population is shown in table 1. Treatments with schradan were compared with other aphicides and the calico scale population increased with the number of years that schradan was applied. The increase continued into 1957 and the plots treated with schradan could be distinguished from the other treatments by a higher incidence of the calico scale.

Why use of the systemic insecticides resulted in an increase of soft scales is not known. Some physiological or physical action might be involved or the chemicals may in one way or another interfere with parasitism. It is possible that the scales that survive the treatments take up sufficient amounts of the systemic materials to become toxic to many of the developing parasites.

CONTROL.—Where possible, control of soft scales should be left to their natural enemies. Of these, parasites are the most important, although Low (1935) and Michel-

Table 2.—Comparative effectiveness of several treatments applied in late fall of 1956 against soft scales infesting walnut at Linden, Calif.

Treatment and Amount Applied per Acre	Date Applied	Average Number of Individuals per Twig Sample: Pre-Treatment[a] Frosted Scale and European Lecanium Complex	Pre-Treatment[a] Calico Scale	Post-Treatment[b] Frosted Scale and European Lecanium Complex	Post-Treatment[b] Calico Scale
25% parathion W.P., 4 lb.[c]	Nov. 7	189.0	12.0	0.66	0.45
25% parathion W.P., 5 lb.[d]	Nov. 7	121.0	5.6	0.43	0.23
25% parathion W.P., 5 lb.+ oil emulsion 1.6 gallons[d]	Nov. 23	107.0	1.7	0.02	0.01
25% parathion W.P., 5 lb.+ oil emulsion 8 gallons[e]	Dec. 17	2.5	17.0	0.00	0.02
10% Phostex+emulsifier 2%+ dormant oil 88%, 5 gallon[e]	Dec. 17	34.0	21.5	0.36	0.40
80% dormant oil emulsion[d]	Dec. 17	20.7	9.2	0.05	0.09

[a] Average number of individuals found on the basal 2 inches of the twig growth of the preceding season.
[b] Average number of individuals on 2 inches of twig growth calculated from the number found on 10 inches of twig growth.
[c] Applied in approximately 83 gallons of water with an air carrier sprayer.
[d] Applied in approximately 100 gallons of water with an air carrier sprayer.
[e] Applied in approximately 200 gallons of water with an air carrier sprayer.

Table 3.—Suppressive action of several aphicides upon the soft scale population on walnut in the experimental orchard at Linden, Calif.

Aphicide	Dates Applied and Amount per Acre	Average Number of Scales per Leaf Sample[a] on June 18		July 20		October 7	
		Frosted Scale and European Lecanium Complex	Calico Scale	Frosted Scale and European Lecanium Complex	Calico Scale	Frosted Scale and European Lecanium Complex	Calico Scale
Demeton (actual)	Apr. 16, 0.5 lb.[b]	1.05	0.49	0.00	0.01	0.01	0.03
	Apr. 16, 0.25 lb.[b]	1.71	1.55	0.06	0.01	0.01	0.05
Schradan (actual)	Apr. 16, 0.5 lb.[b]	2.21	2.86	0.02	0.00	0.01	0.01
	Apr. 16, 1.0 lb.[b]	0.31	1.66	0.01	0.00	0.00	0.01
25% parathion W.P.	Apr. 26, 1 lb.	0.29	0.79	0.05	0.02	0.03	0.13
	June 26, 1 lb.						
25% malathion W.P.	Apr. 26, 3 lb.	1.50	1.00	0.07	0.37	0.20	0.03
	June 26, 3 lb.						
15% guthion W.P.	Apr. 23, 6.64 lb.	0.62	0.44	0.13	0.01	0.07	0.00
	June 21, 5.6 lb.						
Trithion (4 lb./gal.)	Apr. 16, 1 qt.	4.00	0.11	0.00	0.32	0.12	0.00
	June 26, 1 qt.						
25% ethion W.P.	Apr. 26, 10 lb.	0.17	0.21	0.00	0.00	0.00	0.00
	June 21, 15 lb.						
25% Thiodan W.P.	Apr. 26, 10 lb.	0.48	1.57	1.35	0.23	0.05	1.01
	June 21, 15 lb.						

[a] Samples 15 mm. in diameter.
[b] On June 26, plots treated with 2 lbs 25% parathion W.P. and three-fourths gallon of light summer oil emulsion per acre to control soft scales.

bacher & Hitchcock (1957) have shown that the Audubon warbler plays an important role in the control of the calico scale.

Because of serious interference with natural control by certain treatments, insecticidal measures must frequently be relied upon. Investigations have shown that there are two periods of the year when the soft scales can be controlled. One of these is during the summer and the other is from the time the walnut trees drop their leaves in the fall until just before the scales molt and begin rapid growth in late winter and early spring. Treatments after this time and until the eggs have hatched are ineffective even though excessive amounts of insecticides are applied. Summer treatments are best applied during July and August, and winter treatments from about the first of December to about February 20.

Parathion has proved to be one of the most effective materials tested. The results of a series of experiments conducted in the fall of 1956 at Linden, California are given in table 2. Where parathion was used alone there was a marked reduction in the scale population. The control was very much improved where parathion was used in combination with a light medium summer or a dormant oil emulsion. The principal reason for this, as also shown by previous experiments, was that parathion alone at the dosage used was not effective against the European fruit lecanium nor was it very effective against the calico scale. Use of oil alone resulted in satisfactory control. Phostex, at the dosage used, gave a little below an effective level of control

In experimental plots at San Jose and Walnut Creek satisfactory control of the scales was obtained with approximately 5 pounds of 25% parathion wettable powder, plus 4 gallons of oil emulsion per acre, applied with air carrier sprayers in from 150 to 200 gallons of water per acre. In an experimental orchard at San Jose promising results were obtained with Dinitro 289 (36% active) at the rate of 4 gallons per acre in 165 gallons of water. It is well to point out also that the winter treatments with parathion resulted in an excellent kill of the overwintering eggs of the walnut aphid.

The scales were more easily killed in the summer than in the winter. Results of certain treatments against the walnut aphid and their effect upon the soft scale population in the experimental orchard at Linden are given in table 3. Parathion, 25%, wettable powder which had been used on the same plots over a period of years at a rate of 1 pound per acre resulted in keeping the scale population below a destructive level. Three pounds of 25% malathion wettable powder also gave sufficient protection. Plots treated with demeton or schradan needed treatment for scale control. On June 26 these plots were treated with 25% parathion wettable powder at the rate of 2 pounds, plus three-fourths gallon of light summer oil emulsion in 100 gallons of water per acre, applied with an air carrier sprayer. This treatment resulted in excellent control of the scales. Ethion, Guthion, and Trithion all showed much promise in controlling the scales.

ACKNOWLEDGMENTS.—We would like to express our appreciation and thanks to C. C. Anderson, Ralph Bishop, H. E. Minahan, H. S. Hight, Wayne Haas and the several growers for their excellent cooperation in the conduct of the investigation. Thanks are also due the firms which furnished insecticides and other assistance. For field and laboratory help we wish to thank the following: Fred Charles, Milton Bell, Don Rough, H. T. Gotelli, Donald Burdick and Marius Wasbauer.

REFERENCES CITED

Bartlett, Blair R., and J. C. Ortega. 1952. Relation between natural enemies and DDT induced increases in frosted scale and other pests of walnuts. Jour. Econ. Ent. **45**(5): 783–5.

Low, H. J., 1935. Birds control calico scale. Pacific Rural Press 130(26): 663.

Michelbacher, A. E., 1955. Frosted scale on walnuts in northern California. Pan-Pacific Ent. 31(3): 139–48.

Michelbacher, A. E., and Stephen Hitchcock. 1957a. Calico scale, *Lecanium cerasorum* Cockerell on walnuts. Pan-Pacific Ent. 33(1): 15–18.

Michelbacher, A. E., and Stephen Hitchcock. 1957b. Soft scales on walnut in 1956. California Agric. 11(3): 7–8, 14.

Michelbacher, A. E., Clark Swanson, and W. W. Middlekauff. 1946. Increase in the population of *Lecanium pruinosum* on English walnuts following applications of DDT spray. Jour. Econ. Ent. 39(6): 812–3.

Middlekauff, W. W., A. E. Michelbacher, and Clark Swanson. 1947. Increase of frosted scale following use of DDT and other sprays. Jour. Econ. Ent. 40(3): 442–4.

Serr, E. F., 1933. Calico scale—a new pest and how to control it. Diamond Walnut News 15(4): 9.

Control of the Cyclamen Mite on *Jasminum sambac* (L.) Ait. in Hawaii[1]

FRANK H. HARAMOTO and W. WAYNE BOYLE,[2] *Hawaii Agricultural Experiment Station, University of Hawaii, Honolulu*

ABSTRACT

During the spring of 1956, fields of pikake, *Jasminum sambac* (L.) Aitken, in Honolulu were so badly infested (about 90% of the shoots) with cyclamen mite, *Steneotarsonemus pallidus* (Banks), that there was almost no production of leis from this flower crop. In chemical control experiments conducted during March and April, three spray materials—10% endrin wettable powder (WP) at 2 lb. per 100 gal., 25% parathion WP at 2 lb. per 100 gal., and Metacide emulsifiable concentrate (EC) at 2 qt. per 100 gal.—effected 40 to 50% control over the check on a basis of infested shoots, with concentrations of one-half this amount for each material giving poorer control. However, parathion and Metacide produced severe phytotoxicity at each concentration, while endrin produced none. During July, endrin EC became available locally, and growers were urged to apply this material at 1 pt. per 100 gal. every 2 to 3 weeks. Complete control of the mite was obtained within 2 months. The continued use of endrin EC has apparently exterminated the mite in all commercial plantings, as infested shoots cannot be found there at present (September 1957). Any ecological factors that may have contributed to the mite's disappearance remain unknown.

The occurrence of the cyclamen mite, *Steneotarsonemus pallidus* (Banks), on pikake, *Jasminum sambac* (L.) Ait., in Hawaii was noted as a new host record for the mite and reported before the Hawaiian Entomological Society in May 1956 (Boyle 1957). Experiments in chemical control of the mite on pikake were done by the authors during the spring of 1956. Preliminary results of these experiments and a discussion of the problem were presented in a short, illustrated paper (Boyle & Haramoto 1956). The object of the present paper is to summarize the entire project and subsequent developments now that the mite appears to be under complete control.

Hawaii residents know pikake as one of the more desirable flowers used in making leis and, to some extent, perfumes. The flower is small (scarcely an inch in diameter when fully opened), white, and very fragrant. Pikake leis are much in demand and command premium prices in comparison with vanda orchids, plumerias, and other popular lei flowers. Buds are picked just before they open, strung on lei thread immediately, and kept under refrigeration until displayed for sale. Commercially, pikake plants are cultivated in hedge-like rows in small fields less than an acre in size. All of the fields on Oahu are located within a few blocks of each other in the Kaimuki district of Honolulu near Diamond Head. Fewer than a dozen such commercial plantings occur on Oahu, and pikake cultivation is thus a small facet of Hawaiian agriculture; nevertheless, those families who grow pikake spend much of their time in the enterprise and derive a large part of their income from it.

When the authors were asked to investigate the cyclamen mite on pikake, this pest had caused so much damage in all fields that the production of leis had virtually ceased. The mite was found to live and reproduce only between the imbricated young leaves at the shoot apex, where it fed on the developing leaves and flower buds and caused both to be distorted and abnormally small at maturity. Infested shoots thus were not producing usable flower buds, and approximately 90% of the shoots in all fields were infested. In desperate efforts to control the pest, growers had been applying a variety of materials and mixtures thereof for some time. But their methods were haphazard and no discernible degree of control had been obtained.

MATERIALS AND METHODS.—Examination of the literature on cyclamen mite control suggested that endrin in particular should be tested, for Morishita & Jefferson (1955) had found that this material controlled the mite effectively on saintpaulias and certain other ornamentals. Pritchard (1949) stated that parathion gave considerable

[1] Published with the approval of the Director of the Hawaii Agricultural Experiment Station as Technical Paper No. 409. Accepted for publication January 2, 1958.

[2] The authors wish to thank Dr. Martin Sherman, Associate Entomologist at the Hawaii Agricultural Experiment Station, for advice in designing the experiments, and Messrs. Shigeru Uyehara and Wilbert Watanabe, Honolulu pikake growers, for the use of experimental field plots.

Table 1.—Control of cyclamen mite on pikake in Hawaii, spring 1956.[a]

Formulation and Amount per 100 Gal.[b]	Mean Per Cent Infested Shoots			Mean Number Adult Mites per Infested Shoot		
	April 6	April 19	May 8	April 6	April 19	May 8
Endrin 10%, WP						
2 lb.	57.5	45.0	45.0	5.0	3.4	4.1
1 lb.	75.0	57.5	67.5	11.2	4.3	5.3
Parathion 25%, WP[c]						
2 lb.	60.0	40.0	47.5	11.9	3.4	5.5
1 lb.	80.0	55.0	85.0	11.8	6.9	6.2
Metacide EC[c]						
2 qt.	55.0	62.5	67.5	9.9	2.8	5.1
1 qt.	75.0	52.5	67.5	13.0	7.4	6.2
Check—water and Triton B-1956	90.0	90.0	95.0	10.2	12.4	14.3
L.S.D. at 5% level	23.5	23.9	29.9	NS	5.4	5.5
at 1% level	32.2	32.7	40.9		7.4	7.5

[a] Counts are based on samples of 40 shoots (10 from each of 4 replicates) for each treatment.

[b] Applications were made on March 19, March 27, April 2, April 11, and April 30.

[c] Parathion and Metacide produced obvious phytotoxicity.

control of the mite on ornamentals when used as a wettable powder spray.

In a preliminary field-plot experiment, endrin, parathion, Aramite, malathion, EPN, Metacide, lindane, and toxaphene were applied to 15 feet of row each, at concentrations approximating the upper limits of manufacturers' recommendations for their use on other crops. Sprays were applied on March 6, and counts of mites on shoot samples were made on March 9. Results showed that of these materials only endrin, parathion, and Metacide produced high mortalities and appeared to merit further testing. In addition, the systemic toxicants, schradan and demeton, were tested on infested shoots in the laboratory at this time, but they gave no indication of effectiveness.

A field that had received no treatment for at least a month was selected in which to conduct definitive tests on the effectiveness of endrin, parathion, and Metacide. The field was staked off into four quadrants of seven plots each. This produced a total of 28 plots, each consisting of 12 feet of row. The 28 plots provided four replicates, one in each quadrant, for the check and two concentrations of each material. The seven treatments were assigned at random to the plots in each quadrant.

Seven sprays were made up as follows: 10% endrin wettable powder at 1 and 2 pounds per 100 gallons; 25% parathion wettable powder at 1 and 2 pounds per 100 gallons; Metacide emulsifiable concentrate (active ingredients: 6.6% parathion and 26.7% dimethyl analog of parathion) at 1 and 2 quarts per 100 gallons; and tap water for the check. Triton B-1956 as a surfactant was added to all seven sprays at a volumetric ratio of 1:2000. Application was made with a gasoline engine-powered John Bean sprayer of 5-gallon tank capacity. Four gallons of a given spray were made up at a time, and 1 gallon was applied to each of the four replicate plots receiving that treatment. This amount of spray per plot allowed thorough coverage with some run-off.

During March and April 1956, five applications were made at 1- to 2-week intervals. The dates of application were March 19, March 27, April 2, April 16, and April 30.

Three series of samples were taken to evaluate results of the tests. The first series was taken on April 6, four days after the third application; the second on April 19, three days after the fourth application; and the third on May 8, eight days after the fifth and final application. Each series contained a sample of 10 actively growing vegetative shoots selected at random from each of the 28 plots. The shoots were dissected under a binocular microscope, and for each sample the number of infected shoots and the number of living adult cyclamen mites on each shoot were recorded. Because of the difficulty and inaccuracy involved in doing so, the more numerous immature mites and eggs were not counted; but of course shoots bearing any stage of the mite were termed infested.

Results.—The data are summarized in table 1, and the numbers given there are based on samples of 40 shoots—10 from each of 4 replicates. In terms of economic control, the numbers for mean per cent infested shoots are more meaningful than those for mean number of adult mites per infested shoot, because only uninfested shoots produce usable flower buds. (For example, a sample [A] containing 50% infested shoots would have a greater productivity than another sample [B] containing 60% infested shoots, even though the mean number of adult mites per infested shoot were larger in A than in B.) The data show no significantly better control, in terms of infested shoots, with successive applications of toxicants. Perhaps this is because the first sampling was done after the third application of spray, at which time the toxicants may have already effected their maximum control under the prevailing conditions. The mean numbers of adult mites per infested shoot indicate, however, that the toxicants tended to weaken progressively the mite populations in individual infested shoots in comparison with those in the control plot. In general, the data show that control was better at the higher concentration of each material and that endrin gave as good control as either parathion or Metacide.

From the standpoint of phytotoxicity, endrin was definitely superior to the other materials. Both Metacide and parathion produced plant injury at each concentration, while endrin produced none at either concentration. Symptoms of phytotoxicity—a general yellowing or chlorosis of the foliage and the production of abnormally small, unusable flower buds—were first noticed at the time of the third application and persisted into July, 2 months or more after the final application. Phytotoxicity was more pronounced with Metacide than with parathion and also at the higher concentration of each material. It is not surprising that parathion and Metacide produced similar phytotoxicity, since the active ingredients of Metacide are parathion and its dimethyl analog. Metacide produced a greater degree of phytotoxicity than parathion wettable powder possibly because of being in the liquid formulation.

Subsequent Developments.—On July 16, at a meeting of pikake growers sponsored by the Hawaii Agricultural Extension Service, growers were formally made acquainted with results of the mite-control experiments. At that time, endrin emulsifiable concentrate (containing 1.6 pounds actual per gallon) had just been made available by a Honolulu chemical company, and the authors anticipated a probable increase in effectiveness from the use of this formulation instead of the wettable powder, which was the only form on hand when the tests were

begun. Therefore, it was recommended to growers that they spray with endrin emulsifiable concentrate at the rate of 1 pint per 100 gallons, an amount equivalent to 2 pounds of 10% wettable powder in actual concentration of toxicant.

Results were most gratifying. Within a few weeks growers began reporting satisfactory control and substantial yields of flower buds. It soon became difficult to find infested shoots in the fields and for the past entire year, the cyclamen mite has been under complete economic control in all commercial plantings.

Occasional checks of pikake fields are being made by the authors to keep informed of the mite infestation. A comprehensive check was made on May 27, 1957, when seven fields were thoroughly examined. Each field was searched for shoots that showed symptoms of possible damage by the cyclamen mite, and 10 such shoots were collected from each field. These were dissected under the binocular microscope, and only four shoots of the 70—two from each of two fields—were infested. Most of the other shoots were infested with the long-tailed mealybug, *Pseudococcus adonidum* (L.). On September 13, 1957, six of the same fields (one had been destroyed for residential subdivision) were again examined. This time 20 shoots that appeared possible infested were selected from each field. Not one of the 120 shoots was infested. As before, most of the symptoms suggesting presence of cyclamen mite were caused by the long-tailed mealybug. Thus it appears at the present time (September 1957) that the cyclamen mite has been virtually or entirely exterminated in the pikake fields. However, growers are continuing to spray with endrin every 4 to 8 weeks to keep their fields mite-free—a practice that might profitably be dispensed with.

DISCUSSION.—The question of whether endrin was wholly responsible for the mite's disappearance cannot be answered with certainty. That endrin was in fact the cause of the cyclamen mite's suppression is indicated, first, by the dramatic and sudden control of the mite when growers began spraying with the emulsifiable concentrate and, second, by the lack of any other apparent factor.

However, there was some evidence that unknown ecological factors might have reduced the mite population independently of the use of endrin. For example, one grower, at the request of the authors, set aside a portion of his field to remain unsprayed and thus serve as a check against endrin treatment of all the other commercial acreage. But this plot was not set aside until after it had been sprayed two or three times with endrin emulsifiable concentrate, the last spray having been applied on October 15, 1956, some 3 months after endrin was first recommended to the growers and after the sudden decline in infestation throughout the plantings had occurred. This plot remained untreated for at least 6 months, but no infested shoots were found in it during this period or subsequently. In another case, a small, abandoned pikake field, rather isolated from other fields and surrounded by a tall hibiscus hedge, was discovered by the authors on May 27, 1957, and was one of the seven fields checked on that date for cyclamen mites, as noted above. Neither then nor on September 13, when this field was one of the six checked, did it show evidence of infestation. From what could be learned of its history, the field had been abandoned before endrin was recommended to growers and had never been treated with that material. It is possible of course that this field, being somewhat isolated from the others, had not become infested with the mite in the first place. Moreover, the plants were in such poor condition that perhaps the environment was unsuitable for mite establishment.

Predation does not appear to have been a factor in the suppression of the mite. Only a very few predators of any kind were observed on pikake, and none of these was observed to attack the cyclamen mite. If, however, climatic or other ecological factors were actually in large part responsible for the mite's disappearance, then the infestation can be expected to recur at a future time.

REFERENCES CITED

Boyle, W. Wayne. 1957. Notes and exhibitions. Proc. Hawaiian Ent. Soc. 16(2): 186.

Boyle, W. Wayne, and F. H. Haramoto. 1956. Cyclamen mite on pikake. Hawaii Farm Sci. 5(2): 1, 7, 8.

Morishita, F. S., and R. N. Jefferson. 1955. Endrin for cyclamen mite control on ornamentals. Florists' Rev. 116(3000): 21–23.

Pritchard, A. Earl. 1949. California greenhouse pests and their control. California Agric. Expt. Sta. Bull. 713: 1–71.

Effect of Curly Top-Infective Beet Leafhoppers on Watermelon Plants in Different Stages of Development[1]

ORIN A. HILLS, *Entomology Research Division, Agric. Res. Serv., U.S.D.A.*

ABSTRACT

Curly top-infective beet leafhoppers (*Circulifer tenellus* (Baker)) were placed on watermelon plants in different stages of development. Growth was severely retarded when plants were infested in the cotyledon stage but were apparently unaffected when infested in the two-leaf stage. Therefore little damage can be expected from curly top in watermelons except where plants in the cotyledon stage are heavily infested.

The beet leafhopper (*Circulifer tenellus* (Baker)), because of the virus disease curly top which it transmits, is a problem on many vegetable crops west of the Rocky Mountains, including several cucurbits. Hills & Taylor (1949) showed that, although cantaloup plants were stunted by curly top, they often did not develop recognizable symptoms of the disease. Experiments from 1949 to 1951 (Hills & Taylor 1954) showed that the earlier the stage of development in which cantaloup plants were infested with curly top-infective leafhoppers the more severely they were damaged. A high percentage of plants infested in the cotyledon and two-leaf stages were killed. Plants in the four-leaf stage survived but were stunted and the yield was reduced. Plants in the six-leaf stage and those having a runner length of 8 inches were damaged to a lesser degree, but still showed a definite retardation in growth accompanied by reduction in yield.

In 1955 some experiments were conducted at the Phoenix, Ariz., laboratory to determine the effect of feeding curly top-infective beet leafhoppers on watermelons in different stages of development. The plants were of the variety Improved Peacock-Klondike, which is the most

[1] Accepted for publication January 9, 1958.

FIG. 1.—Appearance of watermelon plants after infestation with curly top-infective beet leafhoppers: A, 55 days after infestation in the cotyledon stage; B, 44 days after infestation in the two-leaf stage

common is the Salt River Valley of Arizona. The leafhoppers were from colonies reared on curly top-diseased sugar beets. Preliminary tests were made in the greenhouse during the winter and early spring to be sure these insects were carrying a strain of the virus to which watermelons would be susceptible, and they were followed by field-plot tests in the spring and summer.

GREENHOUSE TESTS.—Watermelon plants were grown in 6-inch flower pots or in deep greenhouse flats. Infective leafhoppers were confined on the leaves for 4 to 6 days by means of glass-vial cages 7/8 inch in diameter and 1 inch tall. In the first test two plants in the cotyledon stage were exposed to six leafhoppers for 6 days; both plants contracted curly top and growth ceased. In a second test two plants in the four-leaf stage were exposed to five and 10 leafhoppers for 4 days; neither plant showed any evidence of curly top. In a third test eight plants in each of these stages were exposed to three leafhoppers for 1 week. The cages were examined three times during the week and dead leafhoppers replaced with live ones. In the third test growth records were kept which consisted of leaf and runner measurements taken weekly for 4 weeks beginning 3 weeks after infestation. These data showed that growth had been affected in 50% of the plants exposed in the cotyledon stage and in only 12.5% of those in the two-leaf stage—in other words, that plants in the cotyledon stage were the more susceptible to curly top. In all these tests some infected plants showed a definite stunting and shortening of the internodes, others only a cessation of growth.

FIELD-PLOT TESTS.—A randomized block experiment was conducted with three treatments and 11 replicates of single-plant plots. Curly top-infected beet leafhoppers were caged on plants in the early cotyledon stage (treatment 1) and in the two-leaf stage (treatment 2), and uninfested plants were used as checks (treatment 3). Cylindrical cloth-covered cages 4 by 8 inches were placed over the plants, and five leafhoppers were introduced into each cage. After 2 days observations were made, and any dead leafhoppers were replaced with live ones. At the end of 4 days the plants were dusted with 5% malathion, and about 6 hours later the cages were removed. After the last cages were removed, the entire planting was dusted with malathion, and the dusting repeated at approximately weekly intervals thereafter to maintain as nearly insect-free conditions as possible. Some of the infested plants developed curly top symptoms. They consisted usually of a shortening of the internodes causing a bushy plant, but the most conspicuous symptom was cessation of growth. Of the plants infested in the cotyledon stage, 64% were measurably affected (fig. 1), but only 9% of the plants infested in the two-leaf stage were affected.

To determine further the effect of the curly top virus on these plants, the length of the runners and the fully extended leaf blades were measured periodically. These data, presented in figure 2 indicate that the cessation of growth was much more pronounced in plants infested in the cotyledon stage. The leafhoppers were placed on these plants on May 23; by June 17 they had significantly shorter runners and leaf blades than the uninfested plants and by June 28 both runners and leaf blades were smaller than in either the uninfested checks or the plants infested later. This difference increased as the season progressed. The leafhoppers were placed on the two-leaf plants on June 1 and, while there was a tendency toward shorter runners and smaller leaves than in the untreated plants, these differences were not significant at any time during the season.

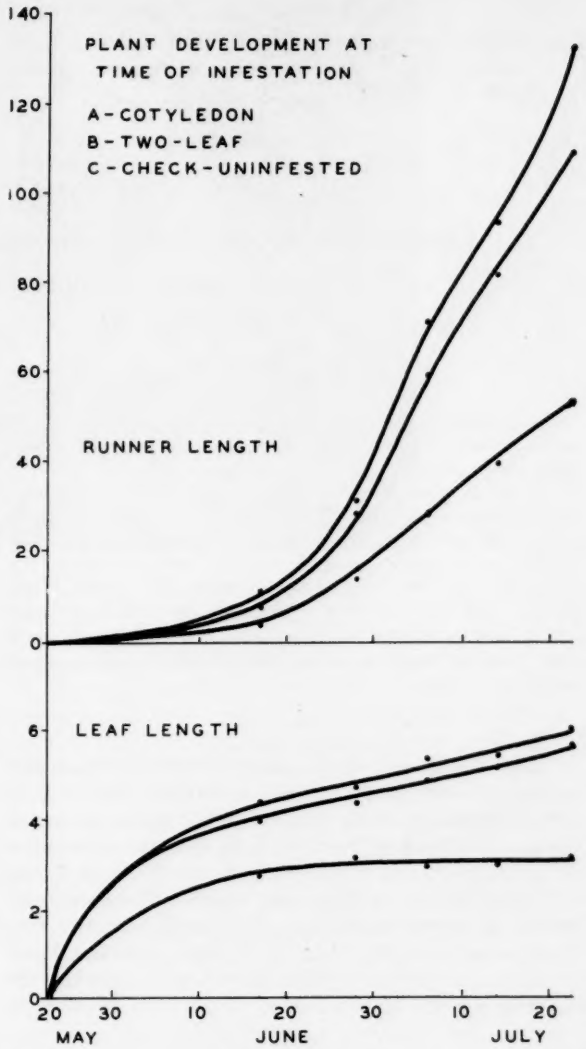


FIG. 2.—Growth rate as indicated by runner and leaf length of watermelon plants infested in the cotyledon and two-leaf stages with curly top-infective leafhoppers.

CONCLUSIONS.—In greenhouse tests watermelon plants in the cotyledon stage were much more susceptible to curly top than plants in the two-leaf stage. Field-plot tests showed a definite retardation of growth when the plants were exposed in the cotyledon stage to curly top-infective leafhoppers, but only a tendency toward growth retardation when they were exposed in the two-leaf stage. Comparison of these experiments with previous experiments on cantaloups indicates that watermelons are not as susceptible to curly top as are cantaloups. Therefore, little damage can be expected from curly top in watermelons except where heavy infestations of beet leafhoppers occur when plants are in the cotyledon stage.

REFERENCES CITED

Hills, Orin A., and Edgar A. Taylor. 1949. Effect of curly top-infective beet leafhoppers on cantaloup plants. Jour. Econ. Ent. 42(6): 897–900.

Hills, Orin A., and Edgar A. Taylor. 1954. Effect of curly top-infective beet leafhoppers on cantaloup plants in varying stages of development. Jour. Econ. Ent. 47(1): 44–46.

Observations on Methyl Bromide Fumigation of Fruit Under Plastic Tarpaulins and at Cold Temperatures[1]

HENRY H. RICHARDSON and HERBERT ROTH,[2] *Entomology Research Division, Agric. Res. Serv., U.S.D.A.*

ABSTRACT

Methyl bromide gas distribution and other tests were made on some commercial fumigations of plums and other fruit stacked under plastic tarpaulins on covered barges, at temperatures near 40° to 45° F. The tarpaulin was an 8-gauge polyvinyl plastic material and was held to the floor of the barge by means of "sand snakes." Where two fans were used, one at one end at the top and one at the other end at the bottom, both pointing to the center, good gas distribution occurred within 30 minutes. When the fans were shut off after 30 minutes the gas concentration held up well for the rest of the exposure. Some small-scale fumigations near 45° to 50° F. indicated plums had a good margin of tolerance to the methyl bromide.

Early in 1955 a series of emergency fumigations with methyl bromide under plastic tarpaulins and at cold temperatures were made by the Plant Quarantine Branch for the treatment at New York City of some imported plums, peaches, and other fruit very lightly infested with a tortricid larva, *Eulia* sp., and a predacious coccinellid beetle, *Eriopis connexa* (Germ.). The fumigations were made on covered barges (fig. 1) tied up at a Hudson River pier. At the request of that Branch some checks were made of various factors having to do with the fumigations, including gas concentration and distribution where fan circulation and temperature varied. Results of these and other related observations are recorded herein, since there appears to be little information available on such types of fumigation, although the studies of Monro (1945) and Brown (1954) are of interest along this line.

PROCEDURES.—The covered barges were first checked for tightness of the flooring, and any cracks were calked and sealed by the commercial operators making the fumigations under Plant Quarantine supervision. The 20-pound lugs of fruit were stacked (approximately 10 to a stack) on wooden-frame pallets in two to four batches on each barge. Each batch consisted of about 180 to 200 stacks and was approximately 35 to 40 feet long by 12 feet wide and 6 feet high, with spacing provided by the regular ¼-inch wooden strips placed on both ends at the top and bottom of each lug of fruit. The pallets were of such a size as to raise all fruit approximately 6 inches above the floor to allow for bottom circulation. Cross wise spaces of about 6 inches width were usually left between every two stacks. The fruit load generally

[1] Accepted for publication January 13, 1958.

[2] Plant Quarantine Division officials R. G. Oakley and W. W. Chapman provided the fruit for some tests, and G. E. Fowler, E. Richman, L. J. Burke, and other plant-quarantine inspectors cooperated in this work.

FIG. 1.—Covered barge tied up beside Pier 57, North River, New York City. A load of fruit being fumigated under plastic tarpaulin is visible through open door at lower center.

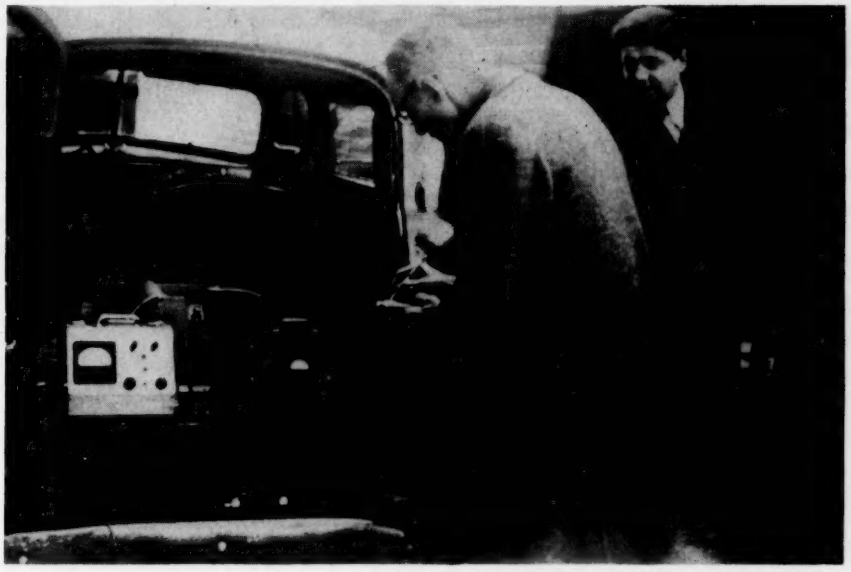


FIG. 2.—Thermal-conductivity gas analyzer in rear of panel truck, placed adjacent to barge (at right) where methyl bromide fumigation of fruit was being made.

occupied about two-thirds of the space under the tarpaulin.

The 8-gauge polyvinyl plastic tarpaulin (35 by 75 feet) was stretched over the fruit and sealed at the floor on all sides with canvas "sand snakes." To permit air circulation the tarpaulin was held 2½ to 3 feet above the load by means of small stacks of fruit boxes located at each top corner and on both sides at the top center; space was also left at the two ends. The tarpaulin was rather stiff and hard to handle at the cold temperatures. A number of rips and tears usually occurred in handling, and it was necessary to patch them with masking tape after each fumigation.

Five to eight ⅛-inch Saran tubes were placed at various locations in the fruit load to determine gas distribution. Analyses of methyl bromide concentrations were made with a thermal-conductivity gas analyzer (fig. 2). From 26 to 32 gas-concentration tests were made in each fumigation. Usually three or more readings were made at 10-minute intervals from the start up to 90 or more minutes of the 2-hour fumigations.

In the first fumigation, at a fruit temperature of about 33° F., a dosage of 4 pounds of methyl bromide per 1000 cubic feet was used. In the three other fumigations at slightly higher temperatures (40° to 46°) 3 to 3½ pounds were used. The dosage was measured by the loss of weight of a cylinder of methyl bromide placed on a scale.

The methyl bromide was applied through a ¼-inch copper tube attached to the cylinder. It was volatilized in a coil of this tubing approximately 20 feet long placed in a pail of hot water. Good vaporization was obtained, as indicated by the fact that the tubing beyond the coil was hot during and shortly after the dosage had been applied. The methyl bromide was ejected into the air stream by the circulation fan at the top front of the load, usually at the rate of 2 to 3 pounds per minute. In the first fumigation only one fan was used and it was run for the entire 2-hour exposure. The fan was an 18-inch, 3-blade pedestal-type, ⅛-hp., 1725 r.p.m., 2.5 amperes, with rated capacity of approximately 3350 cubic feet per minute. In subsequent fumigations another similar fan was placed at the bottom rear end; both fans pointed toward the center of the load. They were run for only the first 30 minutes.

RESULTS OF GAS-CONCENTRATION STUDIES (TABLE 1.)—In the fumigation where only one fan was used at very cold fruit temperatures, gas concentrations were low—mostly near 30% of the original dosage and still not very evenly distributed after 30 to 40 minutes and they fell off to near 20% toward the end of the fumigation. Outside weather conditions were so severe (temperature near 15° F. and winds of about 20 m.p.h.) that it was necessary to use some oil heaters to keep air temperatures up inside the barge. All these conditions probably had something to do with the low and uneven gas recoveries. In the other three fumigations, at higher temperatures and with two fans, gas concentrations came up to near 70 or 80% and were rather evenly distributed in about 30 minutes. After the fans were shut off (30 minutes after the start), the concentrations held up very well for the next 30 minutes and then dropped slowly to about 65 to 70% toward the end of the 2 hours. The irregularity evidenced toward the end of the second fumigation appeared to be due mainly to low recoveries at one particular location; possibly there was a tear in the tarpaulin. Averages for these fumigations were surprisingly uniform. Outside weather was much milder—near 45° to 50°— but windy. Gas analyses also indicated that the methyl bromide penetrated to the center of the fruit package about as fast as it was distributed over the load.

Since separate tests showed that the tissue-paper wrapper on this fruit was permeable to methyl bromide gas, the wrapper did not have to be removed during the fumigation.

On the basis of these results and pending further studies, it was suggested that two circulation fans be used for the first 30 minutes of each fumigation and that

Table 1.—Per cent of original dosage of methyl bromide found at various intervals and locations during four cold-temperature fumigations under plastic tarpaulins.

Minutes After Start	One Fan 2 Hours 33° F.		Two Fans First 30 Minutes					
			45° F.		46° F.		40° F.	
	Range	Average	Range	Average	Range	Average	Range	Average
5– 10	—	—	7–129	60.8	58–75	69.3	63–88	77.8
11– 20	22–47	33.0	7–109	69.5	24–85	63.0	75–79	78.0
21– 30	31–31[a]	31.0	89– 96	92.8	60–75	68.6	67–79	74.0
31– 40	19–38	27.2	79– 88	85.0	67–79	71.6	75–79	76.5
41– 50	22–33	25.6	86.0	[b]	79.0	[b]	71.0	[b]
51– 60	20–28	23.3	77– 91	84.0	68–79	75.3	75–75	75.0
61– 70	15–25	20.6	59– 86	72.5	68–79	74.8	63–75	70.3
71– 80	19–27	21.8	77.0	[b]	71–79	75.6	68–71	70.0
81– 90	20–23	21.6	48.0	[b]	67–79	72.3	63.0	[b]
91–100	19–19	19.0	39– 71	58.8	—	—	—	—
101–110	—	—	62– 75	70.0	—	—	—	—
Weighted average	—	24.6	—	74.2	—	71.2	—	74.2

[a] Values for only two locations taken after about 22 minutes.
[b] Only one sample.

the minimum allowable fumigation temperature be near 40° F.

INSECTICIDE EFFICIENCY.—Few data on the efficiency of the fumigations are available, owing to the very low infestation. Plant-quarantine inspectors intercepted a few tortricid larvae and some coccinellid beetles in the various shipments. The tortricid larvae were found alive, though affected, immediately after fumigation, but died within 24 hours. This is typical of the toxic action of methyl bromide, particularly at cold temperatures. Some of the few coccinellid beetles found before fumigation were alive, but after fumigation all were dead.

TOLERANCE OF FRUIT TO FUMIGATION AT COLD TEMPERATURES.—General observations on the condition of the plums, peaches, and apricots in the commercial shipments indicated no detrimental effect from these cold-temperature fumigations. In addition separate small-scale tests were made on plums, since there had been reports of possible injury to some varieties exposed to methyl bromide fumigation—2 pounds for 2 hours at near 70° F. (Claypool & Vines 1953). Small lots of the President variety of imported plums were exposed to dosages of 3, 4, or 5 pounds per 1000 cubic feet for 2 hours and 4 pounds for 3 hours, all at near 45°, in drum fumigations in the laboratory. The fruit was then stored for 2 to 3 weeks at 50°. Little or no effect on condition or taste of the fruit from any of the fumigations could be detected, confirming the results of some small-scale tests made here in 1954 on cold-temperature fumigation of imported plums. Probably the cold temperature is an important factor in the tolerance results.

REFERENCES CITED

Brown, W. Burns. 1954. Fumigation with methyl bromide under gas-proof sheets. Gt. Brit. Dept. Sci. & Indus. Res., Pest Infest. Res. Bull. 1, 34 pp.

Claypool, L. L., and H. M. Vines. 1953. Commodity treatment of deciduous fruits. Third Special Report on Control of Oriental Fruit Fly, *Dacus dorsalis*, in Hawaiian Islands, published by Senate of State of California, pp. 101–17.

Monro, H. A. U. 1945. Low temperature fumigation. Canadian Entomol. 77: 192–6.

NOTICE TO NON-MEMBER SUBSCRIBERS

Beginning in 1959, subscriptions to the JOURNAL OF ECONOMIC ENTOMOLOGY and to the ANNALS OF THE ENTOMOLOGICAL SOCIETY OF AMERICA will be $15.00 each per year, domestic; $15.60 each per year, foreign. A single order for both publications, if sent to the same address, may be obtained at a COMBINATION PRICE of $27.00 per year, domestic; $28.20 foreign.

Some Aids for Field Photography of Insects[1]

LELAND R. BROWN, *Department of Entomology, University of California, Los Angeles*

ABSTRACT

The construction, calibration and field use of a hand-held bracket uniting a 35-mm. single lens reflex camera and a speedlight lamp (i.e., electronic flash) is described briefly. The bracket permits the intersection of the lens and lamp axes at the subject when the subject is in focus for a continuous range of subject lengths from about 1 m. to 27 mm. Also described are: (a) a remote control device for the lens diaphragm, (b) an eye cup attached to the camera to exclude extraneous light, and (c) a fitted case for the camera, bracket, speedlight and attachments while connected together mechanically and electrically and ready for immediate use.

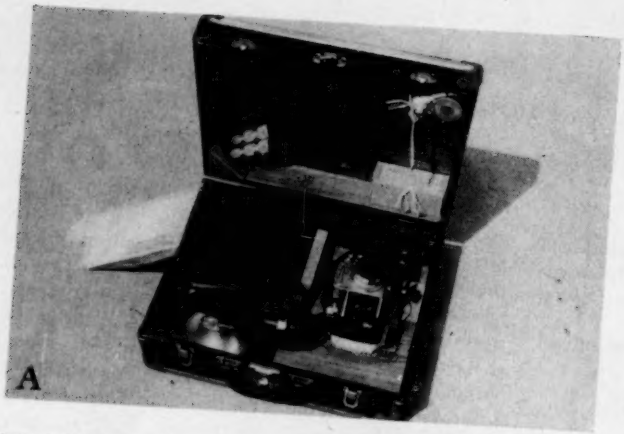
The purpose of this paper is to describe briefly the construction and use of four relatively simple devices which have aided the writer in photographing insects in their natural habitat. These are: (a) a sturdy metal bracket capable of holding the speedlight lamp in a constant, but adjustable, relationship to the camera; (b) a series of three rings and a control cable making possible the pre-setting and rapid remote control of the lens diaphragm; (c) an eye shade or cup that attaches to the camera viewfinder; and (d) a small "suitcase" fitted to receive and protect the camera gear while it is connected together and ready for immediate use. Use of these devices has enabled the writer to place more emphasis on observation of the insect and its activities and less on the mechanical details of obtaining the photographic record, which most entomologists would agree is desirable.

THE BRACKET, ITS CALIBRATION AND USE.—Because of its useful characteristics the speedlight is being employed by an increasing number of insect photographers (Brown 1954, 1955, Pence 1953, Ross 1953). In field photography of insects it is advantageous to have the speedlight lamp and camera connected together as a unit. This usually necessitates some type of connecting bracket. Having the camera and light as a unit offers advantages in carrying on the person, in rapidity and certainty of obtaining the photographic record and in ready storage in some sort of protective case. In spite of these advantages having the camera and lamp together imposes some limitations as compared with a more flexible laboratory set-up where camera and lamp (or lamps) may be separate (Brown 1954, 1955). For a given range of magnifications with the laboratory setup the lamp, independent of the camera, may be positioned at any appropriate distance from the subject and with the light rays striking the subject at a constant angle. With the camera and lamp connected with a bracket for the field, independent control of the lamp distance is not feasible and, as elaborated on below, each magnification will have the light striking the subject at a different angle. Thus calibration methods suitable for a standardized and flexible laboratory camera setup (Brown 1954, 1955) may be only partially suitable for calibrating a field setup.

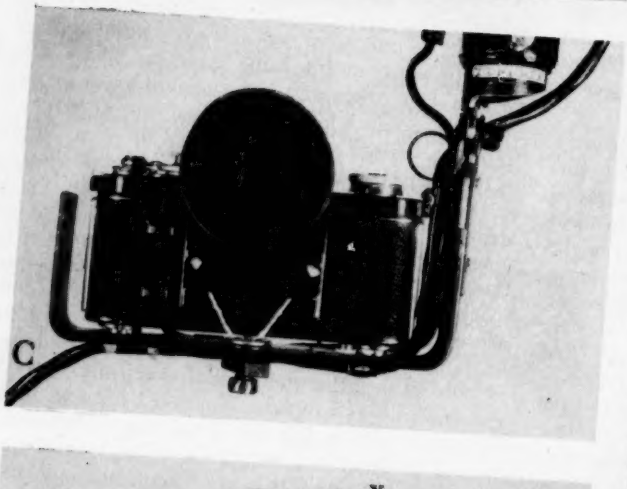
A bracket, as shown in figures A, B, C, E and F, is made from a piece of soft steel, $\frac{1}{8}\times1\frac{1}{4}\times13$ inches, with two right angle, but rounded, bends as shown. The base of the speedlight lamp fits around and is rotated on a piece of $1\frac{1}{2}$ inch outside diameter duraluminum tubing $1\frac{1}{2}$ inches long, and is adjusted thereon by a thumb screw, see top of figure F. A portion of the tubing extends down and is attached to the longest upright of the bracket with a fixed screw and wing-nut. Each millimeter on the circumference of $1\frac{1}{2}$ inch diameter tube equals 3 degrees, approximately. The Exakta camera shown, model V, has an European tripod socket, that is, accepting a $\frac{3}{8}$-16 screw and wing-nut shown attaching camera to bracket; this screw is drilled and tapped in the center for a $\frac{1}{4}$-20 screw or the standard American tripod socket threads. A $\frac{1}{4}$-20 hole is drilled and tapped in the left bracket upright (the shorter upright) for the purpose indicated in figure E. The bracket has a rubber-covered stop near each end of the front of the camera preventing the latter from rotating around the tripod socket. Several small metallic clips are used to hold the electrical cables neatly to the bracket.

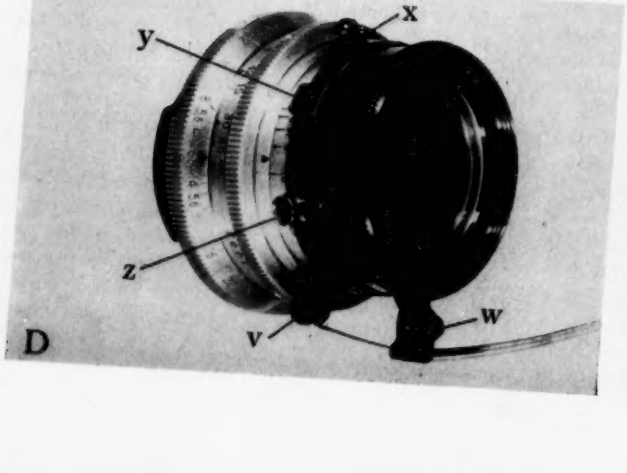
In conventional flash photography, with camera and lamp connected by a conventional bracket, as long as lens and light are pointed in the same general direction and certain simple rules are followed evenly and properly lighted photographs may result. However, as the magnification is increased, that is, the camera approaches the subject as in close-up or insect photography, the illumination will be uneven or will fall off if lamp and lens continue to point in the same direction. With a camera, adjustable bracket and lamp combination as described here and using a white posterboard subject perpendicular to the lens axis, it can be shown that for even illumination of the subject the lamp axis must intersect the lens axis at the subject when the subject is in focus. To demonstrate this it is necessary to select a range of suitable magnifications, such as suggested in column 2 of table 1. It will be noted that in spite of the unusual sequence of magnifications the successive subject lengths, column 1, are at a constant ratio to each other (except 945 and 655), thus assuring an arithmetic and overlapping range of subject lengths and widths. Focusing is by moving camera-bracket-lamp combination back and forth when hand-held or as in figure E by moving the subject. With a lens of 50 mm. focal length these magnifications are obtained as indicated, in a short-hand manner, in column 3 and the footnotes of table 1. The extension tubes are those available from the Exakta Camera Co., which provide a counter ring allowing the engraved lens scales to be turned upright for any amount of extension. Extensions and magnifications indicated in table 1 are in conformity with the formula $X=e/f$, where X is the magnification, e is the amount of lens extension from the infinity position, and f is the focal length of the lens. With a vernier caliper it is desirable to obtain e, without any supplementary extension tubes, corresponding to each mark of the engraved distance scale on the lens (the number in parenthesis in column 3); magnifications in-

[1]Accepted for publication January 10, 1958.

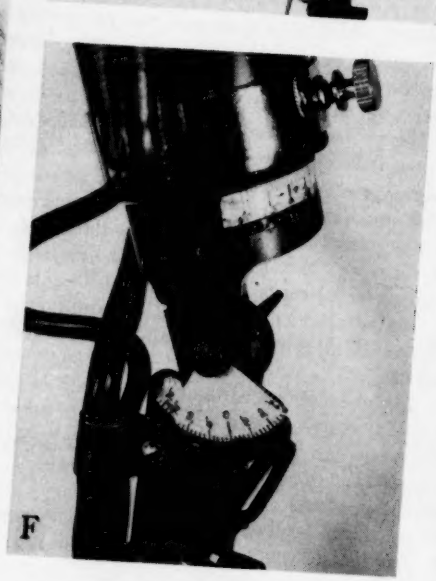

A

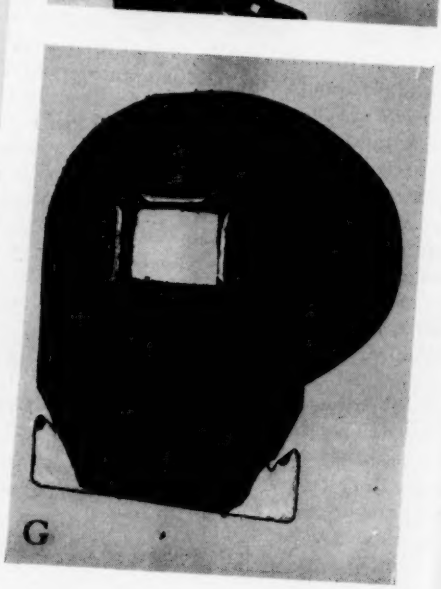
B


C


y
x
z
w
v
D

E


F


G

termediate to those obtainable with the extension tubes alone are then possible with this information.

A simple but accurate method of determining if lens axis and lamp axis intersect at a certain point is as follows: with a strong, continuous light source from behind the camera a subject is brought into focus at a certain magnification. The subject is then replaced by the eye whose reflection in a filter, in a screw-in adaptor ring on the lens, assures that the eye is on the lens axis. By keeping the head in the same position and moving only the eye to peer at the speedlight lamp, the lamp is moved on the adjustable bracket until a reflection of the eye appears in the center of the flat transparent shield on the front of the speedlight lamp; it is assumed then that the eye is on the axis of the lamp. The readings on the lamp adjustment scales are then noted and these form the data in column 4 in table 1, the first number for each magnification being the amount of lamp rotation horizontally (top scale in fig. F) and the second the amount vertically (lower scale in fig. F).

As suggested (Brown 1955) a gray scale is photographed with daylight Kodachrome film at various *f*/no's. (at about 0.18× for a 6-inch gray scale). At the *f*/no. which gives the best reproduction of the gray scale the white subject is photographed at the same magnification to obtain a standard Kodachrome slide. Then the white subject is photographed at various *f*/no's. for the remaining magnifications and that *f*/no. selected which produces a slide most closely approximating the standard slide. The light transmission of the slides may be determined and compared by photoelectric devices (Brown 1955), or much more simply by laying the Kodachromes of the white subject on a white background and viewing and comparing in this manner. Since the light must pass twice through the film in this case small differences in density between slides are readily detectable, even those corresponding to fractional *f*/stops. The *f*/no's. for the given magnifications and lamp orientations are listed in column 5 of table 1. These are for Kodachrome daylight film, with slight correction of an Eastman 81-C filter over the lens, utilizing a Strobonar V speedlight (Heiland Div., Minneapolis-Honeywell, Denver, Colo.). For faster films the same settings are used but neutral density filters (available on special order from the Eastman Kodak Co.) are used over the speedlight lamp, as: ASA 25 films (e.g., Panatomic X)—No. 96 filter (0.40 density), ASA 32 films (e.g., 35 mm. Ektachrome)—No. 96 filter (0.50 density), etc. For those films approximating the same ASA rating as daylight Kodachrome, such as ASA 16 Microfile, no filter need be used over the lamp.

For flat subjects, as documents similar to the white subject above, no additional equipment will be necessary for even and optimum illumination. However, for subjects with some roundness or depth and photographed directly from above a diffuse white reflector, or a mirror, on the side of the lens axis opposite the lamp will usually provide the necessary fill-in illumination. Photographing the subject from the side or as a dorso-lateral view frequently allows the substrate to act as a fill-in reflector.

It should be noted also that the lamp on this bracket, see figure F, can be inclined so as to point toward the ceiling or wall of a room for "bounce or indirect lighting" in conventional photography. As a rule-of-tumb: opening the diaphragm two *f*/stops—from that *f*/no. obtained with a conventional guide number—for a light ceiling of conventional height has provided adequate exposures.

THE DIAPHRAGM CONTROL.—The reader is referred to figure D and its legend. The rings are made from a thin sheet of ferrous metal, those shown being cut from the metal of a film pack. The ring (*v* and *x* in fig. D) on the *f*/no. ring of the lens and the ring (*y* and *z* in fig. D) on the screw-in filter adaptor ring were made of narrow strips of metal which were placed around their respective rings and marked for length, with some overlap, then removed and soldered. The ring (*w* in fig. D), that is gripped tightly between the lens mount and the filter adaptor ring, was made from a piece of flat stock in which a hole was cut large enough to accommodate the male threaded portion

---

FIG. A.—Fitted case with camera, eye cup, bracket, lamp and powerpack in place; lamp has been rotated on mount to fit in case. White area to left is a folded-out and attached plastic sack containing pertinent literature. Note enclosed tripod.

FIG. B.—Camera-bracket-lamp combination in use at 0.18× magnification (refer to table 1) just prior to making exposure—note raised right thumb in ring, i.e., diaphragm stopped down; note manner of holding bracket, and also that it is possible for the operator to wear eye glasses with eye cup. Left index finger is on shutter release. Supporting neckstrap is attached to bracket.

FIG. C.—View of camera, bracket and eye cup from operator's position. Eye cup is secured with a rubber band-loaded knotted string attached around camera tripod socket.

FIG. D.—Illustrates lens mount with diaphragm adjusting mechanism: *v* and *x* are part of a ring attached to rotating *f*/no. ring of lens mount; *y* and *z* are part of a ring which rotates on a supplementary screw-in filter adaptor ring; *w* is an extension of a stationary flat ring which is held securely between the lens mount and screw-in filter adaptor ring. As in figure B, when right thumb is raised remote control cable pulls *v* toward *w* until *x* strikes *y*, thus closing diaphragm; position of *y* is adjusted to proper *f*/no. previous to exposure by set-screw *z*. When thumb is pushed down *v* is pushed from *w* and diaphragm is opened.

FIG. E.—The same as in figure B, but bracket is held by a tripod. Attached to bracket is a rack-and-pinion ("Novoflex, Castel" of Exakta Camera Co.) and attached to that is a rod ("Flexaframe" connectors and rod of Fisher Scientific Co.) and Masonite board, which affords a small, lightweight, smoothly operating and readily adjustable copy stand for the field. Instead of tripod, and with appropriate clamping device, the following could be used: tree, fence post, door, chair, or even the case in figure A.

FIG. F.—Just below lamp set-screw at top is a fixed scale, on 1½ inch diameter tube, for setting horizontal rotation of lamp; white index mark on bottom of lamp opposing scale is visible in figure C. A screw through pivot joint and wing-nut permit adjusting of vertical inclination of lamp as indicated on adjustable quadrant-shaped scale radiating from pivot screw; index mark opposing scale is vertex of light triangular area on adjacent nut. An interval on either scale represents 3°. At 0° on both scales lamp and lens axes are parallel, converging axes being for illumination in close-ups and diverging axes for "bounce" illumination mentioned in text.

FIG. G.—One aspect of the rotatable eye cup or shade illustrating protruding metal "fingers" for attaching to Penta Prism viewer. Eye cup is lined with black velvet as in figure C and as shown here.

Table 1.—Adjustments for field photography of insects with the bracket herein described.

SUBJECT SIZE—MM.	MAGNIFICATION (×)	EXTENSIONS (LENS = 50 MM.)[a]	LAMP ORIENTATION[b]	f/NO.
945×630	0.038×	(4.5′)	2°/ 7°	f/8
655×437	0.055×	(3.25′)	7°/12°	f/8
472×315	0.076×	(2.5′)	8°/17°	f/10
360×240	0.100×	(1.75′)	11°/20°	f/15
276×184	0.130×	2/1, (6′)	14°/25°	f/14
204×136	0.176×	2/1, (2.5′)	18°/31°	f/17
151×101	0.238×	♂, ♀, (4.5′)	22°/38°	f/20
114× 76	0.314×	♂, ♀, 5, (12′)	25°/45°	f/20
86× 57	0.418×	♂, ♀, 5, 2/1, (9′)	30°/50°	f/18
65× 44	0.550×	♂, ♀, 15, (3.5′)	36°/58°	f/18
48× 32	0.750×	♂, ♀, 5, 15, 2/1, (3.5′)	38°/60°	f/18
36× 24	1.000×	♂, ♀, 5, 30, (1.75′)	39°/60°	f/18
27× 18	1.330×	♂, ♀, 5, 15, 30, 2/1, (6′)	39°/63°	f/15

[a] Number in parenthesis refers to footage scale engraved on lens mount. The numbers "2/1" refer to the "Two-in-one" adaptor ring (5 mm. extension) for the Exakta camera. The symbol "♂" refers to male bayonet adaptor and "♀" to female bayonet adaptor; ♂ plus ♀ equals 10 mm. The numbers "5," "15," and "30" refer to lengths in millimeters of extension tubes.

[b] The first number represents degrees of horizontal rotation of speedlight lamp (top scale in fig. F) and second number the amount of vertical rotation (lower scale in fig. F); in both cases rotation is toward the lens axis.

of the filter adaptor ring; also portion *w* was allowed on this ring to provide a stationary connection for the flexible control cable. Set-screws *x* and *z* on their respective rings and connections at *v* and *w* were fashioned of size 2-56 brass nuts and screws heavily soldered in place over holes (no. 44 drill) drilled in the ring. The flexible control cable is made of number 6 music wire, there being two essential parts: a movable inner straight piece, which transmits motion from the right thumb to the *f*/no. ring, and a stationary outer coiled piece with one end fastened at *w* in figure D and the other end at the camera neck-strap eyelet on the right, see figure C. Each end of the coiled piece has a small encircling metal tab fastener which is crimped slightly in the center for holding the coiled wire securely. Making the coiled portion can be accomplished by suspending in a drill press chuck a larger diameter wire, for a temporary core, with a suspended weight, like a 5-pound piece of lead. By slightly squeezing the core wire and music wire between two blocks of wood the music wire coils on the core when turning the chuck by hand. Under power there could be some hazard from the stiff music wire. After some practice suitable perfect lengths can be obtained. Although the writer could not find such, the reader may find commercially a suitable shutter release cable which could serve as the remote control cable described here.

For pre-setting set-screw *z* is loosened and *x* and *y* are moved together to the proper *f*/no. indicated on the engraved scale of the lens and set-screw *z* is tightened. Focusing is done with the diaphragm opened and then it is stopped down by raising the right thumb which draws *v* toward *w* until *x* strikes *y*. This device does not close the diaphragm as rapidly as the "automatic" lenses currently available, but both closing and opening are controlled by a simple movement of the thumb. Also with this device any stop, including those intermediate to the engraved stops, can be pre-set; as shown in column 5 of table 1 with this particular combination of equipment only two magnifications are at an engraved *f*/no., the remainder being intermediate stops.

THE EYE CUP OR SHADE.—The reader is referred to figures B, C, E and G and their legends. The cup is made, literally, from a plastic cup, the one shown being supplied normally for use with a 1-pint "Thermos" bottle. As indicated in figures B and E the cup is cut with a hand coping saw to confrom to the individual's contour around the eye, the cup edges being subsequently smoothed and beveled with a knife and sandpaper. Most of the bottom of the cup is removed leaving a large round hole but enough is left to provide about ¼ inch flange. The second piece of the shade is a flat piece of sheet metal, see figures C and G, with a square hole cut and "fingers" bent out to provide a method of attachment to the Penta Prism viewing device of the camera. Four metal clips are soldered to the cup side of the metal, then the hole of the cup fits around these with the clips being bent over the flange of the cup to hold the cup and metal part together. The clips fit loose enough so the cup can be rotated in either direction, for the right or left eye and for vertical or horizontal picture format. The side of the metal toward the camera, see figure G, is lined by gluing on black velvet cloth. The metal visible from inside the cup, see figure C, is covered similarly, the velvet overlapping the cup flange, thus assuring a light-tight seal. To further reduce reflections the cup and exposed metal parts are painted with flat black paint.

The rapidity and accuracy of focus with any ground glass camera is largely dependent upon maintaining as bright an image as possible on the ground glass focusing screen. Any light that enters the eye, other than that passing through the camera lens, serves only to hinder the focusing process to a greater or lesser degree. Also in close-up photography the brightness of the focusing image is reduced, as at 1.0× magnification the image brightness is only 25% of that at infinity. The writer knows of no eye shade available commercially for this camera, unfortunately, that will accomplish the desirable effect of the one described here, especially for the person that must wear eye glasses.

THE PROTECTIVE CASE.—The reader is referred to figure A which illustrates the small fitted case opened to show the camera, eye cup, bracket, speedlight lamp, speedlight powerpack and tripod in place. The equipment shown here, except the tripod, is connected together mechanically and electrically, and is ready for immediate operation, thus there is little setup or take-down loss of time. The case is a "skate case," presumably ice or roller skates, made by the Standard Trunk and Suitcase Co., Los Angeles, Calif., their model "16-M," and available for a very nominal sum. Presumbably such a sturdy, metal-covered wooden case is available in other areas. The internal dimensions are $15\frac{1}{4}\times11\frac{1}{4}\times6$ inches. Appropriate pieces of wood are attached inside of the case, by round headed wood screws from the outside of the case, to position the various pieces of equipment and for attachment of the sturdy slotted metal plate holding the camera tripod screw. A 2-inch thick pad of sponge rubber is glued to the case beneath the camera. It can be noted that a small amount of space remains for the few necessary attachments and other small items, such as extension tubes, exposure meter and film.

REFERENCES CITED

Brown, L. R. 1954. A standardized laboratory apparatus for using the speedlight in photography of insects and other small objects. Lepidopterists' News **7**: 148–66.

Brown, L. R. 1955. Speedlight calibration for photography of small opaque objects. Jour. Econ. Ent. 48: (1): 82–5.
Pence, R. J. 1953. A simple stage for single lens three-dimensional photomacrography. Turtox News 31: (9): 1–4.
Ross, E. S. 1953. Insects close up. 1–81 pp., 101 photographs, numerous diagrams. Univ. of California Press, Berkeley and Los Angeles.

Insecticides and Spray Schedules for the Control of Cabbage Leaf Feeding Lepidoptera[1]

DAN WOLFENBARGER and EDWIN T. HIBBS,[2] *Iowa State College, Ames*

ABSTRACT

Insecticides and schedules of application were evaluated in Iowa for the control of the cabbage leaf-feeding complex of the diamondback moth, *Plutella maculipennis* (Curt.), the imported cabbage worm, *Pieris rapae* (L.), and the cabbage looper, *Trichoplusia ni* (Hbn.), infesting cabbage grown for fresh market sales in July.

Relative amounts of leaf damage caused by the combined feeding of these insects was visually rated in five classes. The data obtained were suited to statistical analysis. The method of evaluation was economical of time.

Endrin, 0.25 pound per acre, was superior to other compounds tested. Guthion, parathion and Thiodan at 0.75, 0.33 and 0.50 pound per acre, respectively, were superior to DDT, 1 pound per acre.

Endrin and parathion at 0.3 pound per acre applied weekly were equally and extremely effective. Phosdrin, 0.3 pound per acre, was nearly as good as endrin. Biweekly applications of DDT, 1 pound per acre, were inferior to weekly applications of DDT at the same rate, or to toxaphene, 2 pounds per acre, and to parathion applied weekly or biweekly. Methoxychlor, 2 pounds per acre, frequently recommended in the North Central States, was of little value in preventing the leaf damage.

Insecticides for the control of leaf-feeding caterpillars infesting cabbage were compared during 1956 and 1957 in the Muscatine, Iowa, area of fine sandy loam along the Mississippi river where cabbage is grown for fresh market sales during July. The diamondback moth, *Plutella maculipennis* (Curt.), is active early in the spring before heading and is tolerated until the spray applications begin for the control of the imported cabbage worm, *Pieris rapae* (L.), in early June. The cabbage looper, *Trichoplusia ni* (Hbn.), first appears in this area about mid-June and steadily increases throughout July. The latter two species were abundant during the growing seasons of 1956 and 1957. In this area as the commercial crop matures it must be protected from damage by these two species from mid-June until harvest. Usually control is needed between the first and second cuttings which may be one or, at most, 2 weeks apart.

Chapman (1954) found that DDT, rotenone, and methoxychlor were most commonly recommended in the North Central States and that a few states suggested parathion or malathion. He stated that usually three applications were needed for control of these pests. This paper reports an evaluation of the present standing of

[1] Journal Paper No. J-3323 of the Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa. Project No. 1103. Accepted for publication January 10, 1958.

[2] Graduate Assistant and Associate Professor, respectively, Department of Zoology and Entomology.

Table 1.—Comparison of effectiveness of emulsifiable formulations of insecticides against leaf-feeding lepidopterous larvae on cabbage. 1956.

INSECTICIDE	POUNDS PER ACRE	MEAN VISUAL RATING[a]
Endrin	0.25	1.08
Guthion	0.75	2.17
Parathion	0.33	2.29
Thiodan	0.50	2.50
DDT	1.00	3.50
Malathion	1.00	3.50
Thimet	1.00	3.71
Demeton	0.50	4.08
Dieldrin	0.25	4.46
Methoxychlor	2.00	4.50
Dipterex[b]	1.00	4.75
Chlorthion	0.75	4.79
Heptachlor	0.50	4.83
Untreated	—	4.96
$D_{.05}$ (Tukey Range Test)		0.96

[a] Based on two ratings of relative damage (1 least, 5 greatest) July 27 and August 9 combined. Means paralleled by the same line are not significantly different.

[b] Soluble powder formulation.

Table 2.—Comparison of effectiveness of emulsifiable formulations of insecticides against leaf-feeding lepidopterous larvae on cabbage. 1957.

INSECTICIDE	POUNDS PER ACRE	MEAN VISUAL RATING[a]
Endrin	0.30	1.05
Parathion	0.33	1.10
Phosdrin	0.30	1.95
Guthion	0.75	2.26
Thiodan	0.50	2.35
Sevin[b]	1.00	2.65
Perthane	1.00	3.00
Dieldrin	0.30	3.60
Untreated	—	4.80
$D_{.05}$ (Tukey Range Test)		0.91

[a] Based on four ratings of relative damage (1 least, 5 greatest) July 11, 18, 23 and 24 combined. Means paralleled by the same line are not significantly different.

[b] N-methyl-1-naphthyl carbamate.

FIG. 1.—Classes of relative leaf damage by the imported cabbage worm and the cabbage looper; (top) class 1, least damage, (middle) class 3, intermediate damage, (bottom) class 5, greatest damage.

DDT and methoxychlor in relation to other recently developed compounds for the control of the damage caused by the imported cabbage worm and the cabbage looper. In addition the relative merits of weekly and biweekly applications were evaluated.

METHODS AND MATERIALS.—In both years treatments were evaluated by use of a visual-rating scheme. Visual ratings of the relative amounts of leaf damage caused by the feeding of both insect species were characterized by five classes, the class of least damage rating 1 and the class of greatest damage rating 5. All plants in each plot were viewed by two observers independently and the mean rating was recorded. Figure 1 illustrates the minimum leaf damage (class 1), intermediate damage (class 3), and the most severe damage (class 5) which occurred in the plots in 1957. Appropriate tests for the distribution of F and the Tukey range test (Snedecor 1956) for differences among treatment means were applied to the data.

In 1956 insecticides for the control of the imported cabbage worm and the cabbage looper were compared in plots two rows wide and 20 feet long, replicated six times. Applications were made with a three-gallon compressed-air sprayer at the rate of 50 gallons per acre with 12 weekly applications starting May 17, four weeks before heading, and ending August 2, at harvest. The results of these evaluations appear in table 1.

In 1957 plots for the comparison of insecticides were four rows wide and 60 feet long, replicated five times. They were sprayed at the rate of 50 gallons per acre with a tractor-mounted row-crop sprayer equipped with one overhead and two lateral nozzles per row especially designed and built with the cooperation of W. G. Lovely of the Department of Agricultural Engineering, Iowa State College, and of the Farm Machinery Section, Agricultural Engineering Research Branch, Agricultural Research Service, United States Department of Agiculture, for use in experimental plots. The first sprays were applied June 22, one week prior to heading. They were repeated weekly until July 17, one week prior to harvest. Five applications were made. Results of these comparisons appear in table 2.

In the comparison of weekly and biweekly applications of DDT, toxaphene, and parathion the plots were eight rows wide by 75 feet long, replicated six times. Perthane was included in the biweekly schedule only. Applications were started June 21 and ended July 16. Results of these evaluations are presented in table 3.

DISCUSSION.—There were no indications of the failure of previously used insecticides to control the cabbage caterpillars in these tests if the compounds were applied on a well-timed schedule. The method of visually rating the relative amount of leaf feeding appeared to be adequate if the purpose of the investigation is merely to compare the efficacies of treatments in the prevention of leaf damage. It is probably more economical of time than are methods in which larvae must be counted. The method permits flexibility in selecting the time when the evaluations may be made since the destruction of leaf

Table 3.—Comparison of effectiveness of emulsifiable formulations of insecticides applied weekly and biweekly against leaf-feeding lepidopterous larvae on cabbage. 1957.

Insecticide	Pounds per Acre	Weekly or Biweekly	Mean Visual Rating[a]
Parathion	0.33	Weekly	1.208
DDT	1.00	Weekly	2.357
Parathion	0.33	Biweekly	2.587
Toxaphene	2.00	Weekly	2.668
DDT	1.00	Biweekly	3.585
Perthane	1.00	Biweekly	3.585
Toxaphene	2.00	Biweekly	3.710
Untreated	—	—	4.857
$D_{.05}$ (Tukey Range Test)			0.409

[a] Based on four ratings of relative damage (1 least, 5 greatest) July 11, 18, 23 and 24 combined. Means paralleled by the same line are not significantly different.

tissues by feeding larvae provides a record of damage that is persistent throughout the season.

That endrin was superior to other compounds in the prevention of leaf damage is apparent from an examination of the data presented in the tables. DDT provided adequate control if applied frequently. Methoxychlor was of relatively little value in preventing the damage.

Among the phosphate insecticides, parathion was superior and Guthion was promising. Since applications may be needed for control of leaf feeding insects near harvest time, Phosdrin will be useful in preventing the damage at that time.

References Cited

Chapman, R. K. 1954. Present status and control recommendations for cruciferous insects. Ent. Soc. Amer., North Central States. Branch Proc. 9: 88–91.

Snedecor, George W. 1956. Statistical methods. 5th ed. Ames, Iowa. Iowa State College Press.

Carbamoyl Alkyl Phosphorodithioates as Chemotherapeutic Agents: Effects of Dimethoate against Grubs in Cattle[1]

Redginal Hewitt, John Emro, Joseph Entwistle, John Pankavich, Ralph Thorson, Wyeth Wallace and Emanuel Waletzky, *Research Division, American Cyanamid Company, Pearl River, N. Y.*

Abstract

In 21 cattle, single oral doses of from 2.5 to 40 mg./kg. and intramuscular doses of 5 and 10 mg./kg. of dimethoate (Am. Cyanamid 12,880): O,O-dimethyl-S-(N-methylcarbamoylmethyl) phosphorodithioate killed 86% to 100% of second instar *Hypoderma* larvae in the backs of cattle within 1 week after treatment. The lowest dose tested with *H. bovis* was 5 mg./kg. Ten mg./kg. was the lowest dose uniformly highly effective against third instar larvae.

Single oral or intramuscular doses of 10 and 20 mg./kg. of dimethoate killed 50% or more of the first instar larvae of *H. lineatum* (De Vill.) in the gullets of five calves in total within 14 to 25 days after treatment. A similar effect was obtained in one calf treated with 5 mg./kg. orally and sacrificed 25 days later, but not in two calves treated with 5 mg./kg. intramuscularly.

In calves weighing 118 to 427 kg., single oral doses of 40 mg./kg. and above, of dimethoate produced severe toxic symptoms, and 80 or 100 mg./kg. were lethal in 2 days or less; 15 and 20 mg./kg. produced mild toxic symptoms, and doses lower than 15 mg./kg. were essentially asymptomatic. Data on toxicity in sheep and other animals are presented. No differences were observed between the toxicity of 99% and 94% material, and two different drench formulations in ruminants gave comparable results.

Comparisons are given with other types of compounds that are active chemotherapeutically against cattle grubs.

Following the preliminary screening and evaluations by Hewitt *et al.* (1958) of carbamoyl alkyl phosphates as chemotherapeutic agents in mammalian-mosquito (*Aedes aegypti* (L.)) test systems, Am. Cyanamid 12,880: O,O-dimethyl-S-(N-methylcarbamoylmethyl) phosphorodithioate, or dimethoate, was selected for trials against the grubs, *Hypoderma lineatum* (De Vill.) and *H. bovis* (L.), in cattle. The results of mosquito feedings on sheep and cattle treated with this compound indicated activity comparable to that obtained in rodents, and limited toxicity data from cattle suggested a reasonable margin of safety within dosage limits known to produce aedicidal effects. The present report concerns results obtained in grub-infected cattle observed under laboratory conditions. Field studies will be reported elsewhere (Johnson *et al.*, unpublished data).

Crystalline samples of dimethoate of 99% purity, obtained from Dr. R. W. Young, Agricultural Chemicals Section, Research Division, American Cyanamid Company, were used throughout except where noted otherwise.

Methods.—*Studies on second and third instar larvae.*—Cattle infected with *H. lineatum* and/or *H. bovis* were obtained from several sources. Dairy cattle were purchased on the local (New York State) market, and Herefords were from Montana and South Dakota.

The number of grubs at the time of treatment (late December and early January for the *H. lineatum* animals, and principally March and April for those with *H. bovis*) varied from seven to greater than 100, including both second and third instars in most cases. Four out of eight examined from late March through April had first

[1] Accepted for publication May 2, 1958. Partial cost of publication of this paper was met by the Research Division of the American Cyanamid Company. Pearl River, N. Y.

Table 1.—The effect of dimethoate administered internally against second and third instar larvae of *Hypoderma* in calves.

Calf No. and (Type)[a]	Weight (Kg.)	Single Dose (Mg./(Kg.)[b]	Per Cent *Aedes* Killed (No. Fed)	Per Cent Dead Grubs (No. Present) at Sacrifice 6 to 8 Days After Treatment		Species of Grubs[c]
				2nd Instar	3rd Instar	
Oral dosage						
44 (H)	182	40	No data	100 (9)	100 (68)	L
31 (H)	254	40	No data	100 (1)	100 (28)	L
86 (D)	145	40	No data	93 (41)[d]	None found	B
57 (H)	264	15	82 (11)	None found	86 (7)	B
34 (H)	191	10	21 (19)[e]	100 (37)	100 (56)	L
49 (H)	246	10	91 (13)[e]	100 (5)	100 (28)	L
45 (H)	218	10	100 (9)	100 (3)	67 (9)	L
39 (H)	191	10	100 (13)	100 (9)	84 (19)	L
47 (H)	209	10	80 (14)	100 (10)	76 (26)	L
70 (H)	245	10	No data	100 (4)	90 (10)	B
36 (H)	237	5	66 (15)[e]	100 (36)	100 (18)	L
42 (H)	200	5	70 (23)[e]	100 (5)	12 (17)	L
44 (D)	191	5	10 (19)	90 (26)[f]	None found	B
65 (H)	227	5	No data	100 (2)	20 (5)	B
30 (H)	227	2.5	10 (19)[e]	86 (29)	6 (36)	L
35 (H)	227	2.5	32 (10)[e]	94 (55)	38 (21)	L
37 (H)	200	1.0	0 (21)	0 (1)	18 (11)	L
Intramuscular dosage						
40 (H)	264	10	96 (26)	100 (1)	86 (21)	L
67 (H)	200	10	100 (7)	100 (3)	100 (9)	B
54 (H)	227	5	100 (23)	100 (1)	70 (10)	B
72 (D)	109	5	No data	None found	35 (20)	B

[a] H = Hereford; D = Dairy.
[b] Oral doses given by drench in 0.2% agar, except in calf 57 treated with a bolus.
[c] Species based on 50% or more examined: L = *H. lineatum;* B = *H. bovis.*
[d] All of 12 first instars alive in neural canal.
[e] 24 hours after feeding; all others, 72 hours after feeding.
[f] Four of 18 first instars dead in neural canal.

instars in the neural canal, but none in the gullet; all of these had *H. bovis* only in their backs.

Three or four grubs each were removed from most of the cattle prior to treatment. Of these, 61/63 were alive. Cattle treated as indicated in table 1 were held for about 1 week after dosage. At least daily observations for overt symptoms were made, and occasionally a few grubs were removed before the end of the 1-week holding period. Most of the grubs were not disturbed until 6 to 8 days after treatment, when the animal was either killed by a pistol shot and the grubs removed from the stripped hide, or, in a few cases, grubs were removed from the living animal.

The criteria of death of the grubs were 1) lack of movement in saline or water under a low-power dissecting microscope lens, 2) shrinkage and distortion, with partial disintegration, and 3) nearly complete disintegration with only remnants of characteristic structures discernible.

Species and instar differentiations were made according to the descriptions of Laake (1921), James (1947), and Scharff (1950). Usually, at least 50% of the grubs from each animal were classified according to species.

Methods for rearing *Aedes aegypti* and feeding them on mammals are described in a preceding paper (Hewitt *et al.* 1958). Data from mosquito feedings are included in table 1 to illustrate comparative activity against *Aedes* and *Hypoderma*.

Oral doses of dimethoate were administered by drench, with the exception of three cattle treated with boluses. The volume was usually 100 cc. of a mixture of one part acetone or ethyl alcohol to nine parts of 0.2% agar in water. The material was first dissolved in the alcohol without heating. Intramuscular injections were given in volumes of 3 to 10 cc. In one calf (table 1), the intramuscular menstruum was 0.2% agar in water with 10% acetone; in three others, diethyl succinate was used as a solvent (35% solutions). (For other types of formulations used in toxicity studies, see section on results.)

Table 2.—The effect of single oral and intramuscular doses of dimethoate on first instar *Hypoderma lineatum*.

Days After Treatment	Per Cent Dead (Total No. 1st Instars Found)[a]					
	Mg./Kg. Orally			Mg./Kg. Intramuscularly		
	20	10	5	20	10	5
2	0 (9)[b]					
3	0 (1)					
4	0 (3)[b]		0 (34)[c]	0 (5)[b]		
7	14 (14)[b]	9 (21)	—	—		
14	84 (31)	67 (40)	18 (17)	—		
16		100 (17)	—	50 (8)[c]	41 (35)	
19			0 (11)[c]	—	—	
25			60 (24)[c]	—	54 (26)[d]	
28						0 (35)
37						0 (13)

[a] One calf each except in [d]. 84% of grubs in the gullets.
[b] More than 50% of the live grubs abnormal in appearance.
[c] Eight to 25% of the live grubs abnormal in appearance.
[d] Total from 2 calves with 75% and 20% dead of 16 and 10 grubs, respectively.
[e] Treated July 25, all others June 17.

First instar totals in 10 untreated calves sacrificed at various intervals before and during the above experiments were 11, 12, 18, 19, 20, 20, 23, 42, 47 and 73, respectively; all were living and normal in appearance. 99% pure dimethoate was used throughout.

Studies on first instar larvae.—Hereford calves (table 2) were selected in May, 1957 from a herd in Texas and shipped to Pearl River, N. Y. Four of these animals were slaughtered in Texas, and revealed 12, 18, 19 and 42 first instar larvae, respectively, in their gullets. A fifth had 10 larvae in the one-half of a gullet examined. Eighteen calves were treated with dimethoate about 3 weeks later (June 17th) and one about 7 weeks later (July 25th). Four calves held until grubs appeared in the backs had no noticeable warbles until late October, indicating that the treatments with dimethoate (table 2) were given 3 to 4 months before the grubs would have reached and penetrated the hides.

The calves were killed by a pistol shot from 2 to 37 days after treatment and the viscera were examined for the presence of first instar larvae. Five untreated calves were sacrificed at various intervals throughout these experiments and had from 11 to 73 living first instar larvae (see table 2), chiefly in the submucosa of the gullet. "Normal" larvae were motile when placed in warm saline solution, were translucent, and had clearly-defined internal structure. "Abnormal" live larvae from treated calves were sluggish, cloudy in appearance, and the mid-gut was shrunken or distorted. Most of the dead larvae and some of the abnormal, live larvae from treated calves were encased in tough, fibrous "capsules"; the former were partially or nearly completely disintegrated. Blood had sometimes infiltrated the capsules and the appearance of the area suggested an inflammatory reaction. None of 174 first instar larvae removed from five untreated calves during the course of these experiments were abnormal in appearance or were encased in fibrous capsules.

Toxicity.—Acute toxicity studies were made on normal mice, dogs, sheep, and cattle, in addition to all the cattle

with grubs. Mice were treated orally by tubing; dogs by gelatin capsules. Lethality within 3 days was the sole criterion of toxicity used for mice. In the larger mammals, observations for overt symptoms were made for at least 5 days after treatment. The formulations used to treat ruminants are described under "Results."

RESULTS.—*Activity on second and third instar larvae.*—Single oral doses of 5 mg./kg. or more, killed 100%, and 2.5 mg./kg. killed more than 85% of the second instar *H. lineatum* in the backs within 1 week after treatment (table 1). Similarly, 5 mg./kg. orally, or more, killed 90 to 100% of the second instar *H. bovis*. The numbers of second instars of both species present in the backs of calves treated with intramuscular doses were too few to determine percentage efficacy. Doses lower than 10 mg./kg. were less uniformly effective against third instar larvae; but at 10 mg./kg. and above, effects against second and third instars were similar. Data on deaths of *Aedes aegypti* mosquitoes fed on the treated cattle 1 hour after dosage were, in general, roughly comparable to the results on the grubs.

Activity on first instar larvae.—Fifty per cent or more of the first instar larvae were dead in at least one calf each treated with 10 or 20 mg./kg. of dimethoate, orally or intramuscularly, when the animals were sacrificed from 14 to 25 days after a single dose (table 2). A similar effect was obtained in one calf after 5 mg./kg. orally sacrificed on the 25th day, but not in two calves treated with 5 mg./kg. intramuscularly and sacrificed on the 28th and 37th days, respectively.

Abnormal, but unencapsulated larvae were found as early as the second day after treatment with 20 mg./kg. orally, as well as at various periods thereafter with this and other doses. Abnormal, live larvae within fibrous capsules were found as early as the seventh day after treatment, and encapsulated dead larvae were more frequent on the 14th day or thereafter. Although the number of calves treated with any one dose and sacrificed at various periods thereafter was limited, the data indicate a progressive increase in the number of dead larvae found from the seventh day on, particularly with the 10 and 20 mg./kg. oral doses. It will be noted that the numbers of grubs in the treated calves were, on the whole, not different from those in untreated calves from the same herd.

Toxicity.—The LD_{50} of crystalline dimethoate, 99% pure, in mice, is 140 mg./kg. orally, with 95% confidence limits of 124 to 157. The highest dose which does not kill mice is about 100 mg./kg. No overt symptoms occurred in one dog each treated with a single oral dose of 100 mg./kg. and 100 mg./kg. twice in one day, nor in three dogs treated with 50 mg./kg. twice daily for five days (total dose = 500 mg./kg.). A fourth dog treated with 50 mg./kg. twice daily vomited after both doses on the fourth day and treatment was discontinued.

The mild symptoms in sheep and calves noted in tables 3 and 4 included salivation, lachrymation, transitory diarrhea, and anorexia, regardless of their degree. In addition to these, severe symptoms included tremors of the limbs and abdominal muscles, labored breathing, incoordination and prostration, sometimes terminating fatally within 24 to 72 hours. Mild symptoms rarely lasted for more than 24 to 48 hours, and if death did not intervene, recovery from severe symptoms generally began within 48 to 72 hours.

Generally, the symptoms of severe toxicity observed in sheep and calves following dosage with dimethoate above 20 to 25 mg./kg. followed the same pattern.

Samples of both 99% and 94% purity were tested for toxicity in mice, sheep, and calves since a large quantity of the 94% material was prepared for field trials. It was conceivable that impurities in the 94% material might contribute to the toxicity. Similarly, the question arose as to whether different drench formulations might influence absorption and thus affect the degree of toxicity. No difference in toxicity was observed between 99% and 94% material when compared in the same tests in any of the three mammalian hosts used, nor were differences observed in sheep or calves between formulations made with alcohol-agar (one part absolute ethyl alcohol and Am. Cyanamid 12,880 to nine parts 0.2% agar in water; total volume 100 cc.) and xylene-triton (25% 12,880; 75% of a mixture of 65 parts xylene: 10 parts triton X-100; dilute

Table 3.—Acute toxicity of dimethoate in calves and sheep.

	Single Oral Dose[a] (Mg./Kg.)											
	200	100	80	75	60	50	40	25	20	15	10	5
Calves[b]												
Dead[c]/treated	—	2/2	3/3	—	0/3	0/1	0/4	—	0/10	0/9	0/10	0/7
Symptoms—severe		2	3		3	1	4		0	1	0	0
mild									10	8	1	0
none											9	7
Sheep[d]												
Dead[c]/treated	3/3	3/3	—	1/10	—	0/14	—	0/2	—	—	0/3	
Symptoms—severe	3	3[e]		0		2[f]		0			0	
mild				8		1		1			0	
none				2		11		1			3	

All drenched with formulations in one part ethyl alcohol to nine parts of 0.2% agar in water, excepting 4 calves and 10 sheep drenched with xylene-Triton X-100 formulations (see text), and 3 calves treated with boluses. No differences in toxicity were noted between like doses with either the two drench formulations or the boluses.

[a] Used 99% material, excepting 2 calves and 12 sheep treated with 94% material. No differences in toxicity were noted between the two samples (see text).
[b] Mostly Herefords, weighing from 118 to 427 kg. (219 ± 13.9 kg.).
[c] Usually within 24 to 48 hours after dosage.
[d] Mostly lambs, weighing from 25 to 71 kg. (38 ± 2.6 kg.).
[e] One treated with 50 mg./kg. 3 days previously.
[f] One treated with 10, 25, and 50 mg./kg. sequentially, with 1 day's rest between doses.

Table 4.—Toxicity of dimethoate in calves following several treatments, with or without additional treatments with another thiophosphate.

Dairy Calf No.	Days After First Treatment	Weight (Kg.)	Oral Dose Except as Noted (Mg./Kg.) Dimethoate	A Related Phosphorodithioate	Symptoms
14	0	144		150	None
	69		40		Severe
	102	164	10		None
	130	200	16		Mild
	201		(spray—1% in 6 L)		None
	241	264	60		Severe
15	0	190		200	None
	17	182		400	Severe
	60	218	60		Severe
	92	191	10		None
	122	227	15		Mild
16	0	121	25[a]		Mild
	41	118		15	None
	56	145		240	Severe
	98	118	20		Mild
	103	118	80		Severe (fatal)
66	0	209	40		Severe
	21		100[c]		None
	51–57	236	19[d]		Mild

[a] Subcutaneous treatment.
[b] Hereford calf.
[c] 100 cc. of an acetone-water mixture (4:6) rubbed on the abdomen with cheesecloth.
[d] In feed 1 mg./kg. for 2 days, then 2, 5, and 10 mg./kg./day for the following three days—mild toxicity with last dose only.

with water to give the dosage in 1 cc./kg.). The latter formulation was suggested by Dr. R. D. Radeleff of the U.S.D.A. Laboratories, Kerrville, Texas. The 94% material was tested at intervals of 1 week to 6 months after preparation, and the 99% material up to 1 year after preparation, with no observed differences in toxicity.

As demonstrated in table 3, single oral doses of 200 and 100 mg./kg. of dimethoate killed all of three sheep in each test. In 10 sheep treated with 75 mg./kg. orally, the symptoms were mild in eight and absent in two; one of the former died within 96 hours, though only mild overt symptoms were seen. Eleven out of 14 sheep treated with 50 mg./kg. were asymptomatic. Severe toxic symptoms occurred in two sheep in this group, and one of these had previous dosage with 25 and 10 mg./kg., respectively, on two previous days (table 3). In one out of two sheep drenched with 25 mg./kg., and all of three sheep given 10 mg./kg., no symptoms occurred.

Five calves given 80 or 100 mg./kg. of dimethoate (99% material) by drench died within 48 hours. Severe symptoms, but no deaths, occurred in eight calves drenched with 40 to 60 mg./kg. of 99% material. Mild symptoms occurred in 18 and severe symptoms in one treated with 15–20 mg./kg. The mild symptoms included slight salivation, diarrhea, rhinitis and anorexia, generally lasting for not more than 24 hours after dosage. In addition to these symptoms, the one calf with severe symptoms became prostrate 1 hour after dosage and had increased respiration and abdominal distress which lasted for about 24 hours. No symptoms occurred in nine out of 10 calves treated with 10 mg./kg. or in any of seven calves given 5 mg./kg. (99% material). One calf treated with 10 mg./kg. had mild symptoms.

Full details on the depression of erythrocyte cholinesterase activity will be reported elsewhere (Sisson & Roesch, unpublished data), but 40 mg./kg. orally produced cholinesterase inhibition of 71 to 84% and 10 mg./kg. of 13%, with intermediate values at intermediate doses.

A limited number of calves, for which data are not given in table 3, responded similarly to intramuscular dosage at levels comparable to the above. Various menstrua were used, including acetone-water mixtures, diethyl succinate, cotton-seed oil, and peanut oil, and 99% material was used throughout. Two calves treated with 80 mg./kg. died within 48 hours. Severe symptoms occurred in one calf treated with 40 mg./kg. At 20 mg./kg. in two calves, the symptoms were mild. No symptoms occurred in any of 12 calves given 5 to 10 mg./kg. Localized areas of necrosis, along the site of the needle puncture, were observed in two calves examined for such lesions from 1 to 2 weeks after treatment. The extent of the necrosis was negligible.

Reports have been published indicating that organic phosphates cause greater toxicity in cattle on "full feed" (Raun & Herrick 1957). In our tests, all the calves received a finishing ration of rolled oats and timothy hay. Large animals received 6 pounds per day, and small animals, 3 pounds per day of rolled oats, in two feedings. Hay was given in amounts of 8 to 10 pounds per day.

Detailed data from four calves treated with progressively lower doses of dimethoate are given below to illustrate the sequence of toxic symptoms:

Calf No. 16; Dairy; 118 kg. Drenched with 80 mg./kg. (preceded by 10 mg./kg. without symptoms 5 days earlier). Profuse salivation, incoördination, diarrhea, anorexia, labored breathing at 6 hours. Tremors, shallow breathing, prostrate at 24 hours. Died at 26 hours.

Calf No. 15; Dairy; 218 kg. Drenched with 60 mg./kg. Profuse salivation, diarrhea at 3 hours; anorexia shortly thereafter. Difficulty in rising, but no incoördination to exercise at 24 hours; slight salivation and anorexia at 48 hours. Normal at 72 hours.

Calf No. 14; Dairy; 155 kg. Drenched with 40 mg./kg. Lachrymation at 3 hours. Moderate salivation, diarrhea, lachrymation at 6 hours; diarrhea, anorexia, moderate salivation at 24 hours. Normal at 48 hours.

Calf No. 243; Hereford; 273 kg. Drenched with 15 mg./kg. Slight salivation, diarrhea, and anorexia before 18 hours and extending to 22 hours after treatment. Essentially normal at 24 hours.

The effects of three to five treatments with dimethoate over a period of several months, with or without additional treatment with a related dithiophosphate, are given in table 4. Cumulative effects over a period of several weeks were not apparent, and severe or fatal symptoms were produced only by doses of dimethoate that produced similar symptoms in experiments with animals previously unexposed to phosphorodithioates.

DISCUSSION.—The laboratory data obtained with dimethoate in calves indicates an appreciable margin of safety between the lowest dosage which kills first instar *H. lineatum* (5 mg./kg.) or second instar *H. lineatum* and *H. bovis* (2.5 to 5 mg./kg.), and the dose which produces mild toxicity (15 to 20 mg./kg.) or severe, reversible toxicity (40 mg./kg.) in the host animals. The results of extensive field trials now in progress will help to determine more accurately the degree of activity and margin of safety.

Third instar larvae of both *H. lineatum* and *H. bovis* may be considerably more resistant to dimethoate than second instars. It is our impression that most of the third instars which were still living 1 week after treatment with oral doses of 2.5, 5, and 10 mg./kg. (table 1) were mature, deeply-pigmented stages that had already damaged the hides considerably and were about to emerge from the warbles.

Data on effectiveness against first instar larvae of *H. lineatum* are limited, although it appears that the action of dimethoate on this stage is less rapid and possibly less complete than against second instars in the hide. For example, a single oral dose of 10 mg./kg. killed all of 68 second instars in the hides of six calves within 1 week after treatment (table 1). The same dosage in calves with first instars in the gullet killed in one calf each only 9% of 21 at 7 days, 67% of 40 at 14 days, and 100% of 17 first instars 16 days after treatment (table 2). Complete kill of the first instars was not obtained in any of the other calves treated, using various dosages and examining at different periods after treatment up to 37 days, although all observed grubs in two animals receiving 20 mg./kg. were abnormal. Longer holding periods between the time of treatment and the time of sacrifice might have revealed a greater percentage kill with other doses. On the other hand, this stage in the deeper tissues may be somewhat less susceptible to treatment with doses of dimethoate that are nearly completely effective against second instars in the hide. Little previous work of this type has been done with either *H. lineatum* or *H. bovis*, and further exploration will be required to determine its usefulness for predicting activity against first instar larvae under field conditions.

Two of the *H. bovis*-infected calves in table 1 (86 and 44D), treated with 40 and 5 mg./kg., respectively, had a total of 30 first instar larvae in their neural canals 1 week after treatment, and the majority of these were living. In these two animals, 92% of the second instars were dead after treatment. On the other hand, in three calves sacrificed 5 weeks after treatment with 40 mg./kg. in a field trial to be reported elsewhere (Johnson *et al.*, unpublished data) all of 3, 1, and 4 first instars in the neural canals were dead as were a total of 39 of 40 second instars in the hides.

The available data do not allow the selection of a preferred method of treatment with dimethoate. In the four calves treated intramuscularly, the percentage kill of second instars was similar to that obtained with like oral doses (table 1). Intramuscular doses seemed less effective against first instar larvae.

Rough comparisons can be made of the degree of effectiveness of dimethoate against cattle grubs with other compounds reported in the literature. According to McGregor *et al.* (1955), the chlorinated hydrocarbons aldrin, dieldrin and lindane were lethal to yearling Herefords at oral doses of 25 mg./kg. After four multiple subcutaneous treatments with lindane or aldrin at 50 mg./kg. or dieldrin at 25 mg./kg. with 28 days between doses, the majority of *H. lineatum* in the back died at some time after treatment was given. None of these compounds apparently affected first instar larvae. *H. bovis* seemed to be somewhat more resistant to lindane and dieldrin than to aldrin.

McGregor *et al.* (1954) found that single oral doses of 25 mg./kg. of Dipterex, 50 mg./kg. of Chlorthion, and 10 mg./kg. of Diazinon, all organic phosphates, killed most *H. lineatum* which were present in the backs of one cow each at the time of treatment. New grubs reached the back 1 week after treatment with Diazinon. With Dipterex after a dose of 100 mg./kg. in one cow, new grubs reached the back at 4 but not 2 weeks later. There was no evidence that any of the above destroyed the first instars of *H. lineatum* in the gullet.

Recently, two organic phosphates have been described which kill young grubs before they reach the backs of cattle. Dow ET-57 is effective at single doses of approximately 100 mg./kg. orally, against all stages of *H. lineatum* and *H. bovis* (McGregor & Bushland 1957, Roth & Eddy 1957, Adkins 1957). Radeleff & Woodward (1957) state that toxic symptoms in cattle are mild with 125 mg./kg. and become progressively worse as the dosage is increased to 400 mg./kg.

Bayer 21/199, although ineffective against grubs already in the backs at oral doses of 25 mg./kg. (McGregor *et al.* 1954), has shown promise as an effective agent for killing first instar larvae when applied as a 0.75% spray (Brundrett *et al.* 1957).

Dimethoate appears to be more active against grubs in the backs of cattle at the time of treatment than any of the above compounds administered orally with the possible exception of Diazinon. Its margin of safety against grubs is greater by the oral route than the chlorinated hydrocarbons lindane, aldrin, or dieldrin and resembles that of Dow ET-57. Preliminary data indicate that it is also highly effective against the first instar larvae of *H. lineatum*.

No differences in toxicity were observed in mice, sheep, or calves between 99% and 94% material, and in ruminants the two different drench formulations used gave comparable results.

It is of interest that this highly active chemotherapeutic agent was chosen from a mouse-mosquito screening program, and that a considerable correlation occurred from aedicidal activity to effectiveness against grubs in cattle.

REFERENCES CITED

Adkins, Jr., T. R. 1957. Field evaluations of Dow ET-57 as a systemic insecticide for the control of the common cattle grub in Alabama. Jour. Econ. Ent. **50:** 474–6.

Brundrett, H. M., W. S. McGregor and R. C. Bushland. 1957. Systemic cattle grub control with Bayer 21/199 sprays. Agric. Chemicals **12:** 36–38.

Hewitt, R. I., A. F. Brebbia and E. Waletzky. 1958. Carbamoyl alkyl phosphorodithioates as chemotherapeutic agents; aedicidal properties in laboratory mammals, sheep, and cattle. Jour. Econ. Ent. **51**(2): 126–31.

James, M. T. 1947. The flies that cause myiasis in man. U. S. Dept. Agric. Misc. Publ. **631.** 175 pp.

Johnson, W. P., R. I. Hewitt and A. L. Shor. (Unpublished data.) Field trials on the use of dimethoate (CL 12,800), an experimental organic phosphorus compound, against cattle grubs.

Laake, E. W. 1921. Distinguishing characters of the larval stages of the ox-warbles *Hypoderma bovis* and *Hypoderma lineatum*, with description of a new larval stage. Jour. Agric. Res. **21:** 439–57.

McGregor, W. S., R. D. Radeleff, H. V. Claborn and R. C.

Bushland. 1955. Dieldrin, aldrin, and lindane, systemic insecticides against livestock pests. Agric. Chemicals 10: 34–36.

McGregor, W. S., R. D. Radeleff and R. C. Bushland. 1954. Some phosphorus compounds as systemic insecticides against cattle grubs. Jour. Econ. Ent. 47: 465–7.

McGregor, W. S., and R. C. Bushland. 1957. Tests with Dow ET-57 against two species of cattle grubs. Jour. Econ. Ent. 50: 246–9.

Radeleff, R. D., and G. T. Woodward. 1957. Toxicological studies of Dow ET-57 in cattle and sheep. Jour. Econ. Ent. 50: 249–51.

Raun, E. S., and J. B. Herrick. 1957. Feedlot tests of the efficacy of Dow ET-57 (Trolene) for control of cattle grubs. Jour. Amer. Vet. Med. Assoc. 131: 421–4.

Roth, A. R., and G. W. Eddy. 1957. Tests with Dow ET-57 against cattle grubs in Oregon. Jour Econ. Ent. 50: 244–6.

Scharff, D. K. 1950. Cattle grubs. Their biologies, their distribution and experiments in their control. Montana State Coll. Agric. Expt. Sta. Tech. Bull. 471.

Sisson, G. M., and P. Roesch. Unpublished data.

Notes on Thrips of Arizona[1]

F. F. BIBBY,[2] *Arizona Fertilizers, Inc., Phoenix, Arizona*

ABSTRACT

There are 24 species (one probably new) comprising 14 genera of thrips listed, along with hosts on which they were collected in Arizona from 1953–58 by the writer and/or associates (exceptions indicated). The records indicate that *Frankliniella occidentalis* (Pergande) was outstandingly dominant with a wide range of hosts.

Unusual records as pests or potential pests of cotton are those of *Frankliniella gossypiana* Hood, *Kurtomathrips morrilli* (Moulton) and *Scirtothrips* sp. prob. new.

The helpfulness to the writer of a list of insects of the Salt River Valley of Arizona (McKinney 1939) is the incentive for publishing the records below: a list of 24 species of thrips (one probably new) collected in Arizona[3] from 1953 to 1958 by the writer and associates, along with host plants, localities, dates of collection and occasional comments and a few inclusions of other collections and observations, some from out-of-state, all indicated by references.

Aeolothrips duvali Moulton or near

Sweeping mixed planting of alfalfa and barley: Tucson, III-16-55.

Anaphothrips zeae Moulton

Alfalfa: Gilbert, X-12-53.
Baileya multiradiata blooms: Pinal Co., II-2-54.
Johnson grass: Goodyear, XI-19-54.

Chirothrips falsus Priesner

Bermuda grass: Wellton, VIII-21-53, damaging commercial seed crop (L. L. Brimhall); Yuma, X-9-53 (Don McCain).

Chirothrips mexicanus Crawford

Bermuda grass: Yuma Co., in combination with *C. falsus*, a serious pest of Bermuda grass seed industry (Roney 1949).

Erythrothrips arizonae Moulton

Cercidium floridum blooms: Phoenix, IV-28-57; Gila Bend, V-1-55.
Sphaeralcea sp.: Maricopa Co., III-6-54

Frankliniella exigua Hood

Baileya multiradiata: Sacaton, V-4-54.
Psilostrophe Cooperi blooms: Pinal Co., XI-7-54.

Frankliniella gossypiana Hood

Baileya multiradiata blooms: Apache Junction, IV-10-55; Sacaton, V-4-54.
Cercidium floridum blooms: Gila Bend, V-1-55.
Cotton: Casa Grande, V-21-56, causing serious damage by blasting small flower buds, resulting in plants being bare of fruit (R. L. Dezember); Marana, VI-4-56, severe damage, leaving plants bare of fruiting forms (Ken Brinkmeyer); Pinal Co., IV-30-54; V-15-54.
Gossypium Thurberi blooms: Sabino Canyon, IX-16-57.
Grain sorghum: Tempe, XI-14-57.
Mesquite: Eloy, IV-28-54; Willcox, VI-2-54.
Populus Fremontii galls: Pinal Co., IV-22-55.
Solanum elaeagnifolium foliage: Marana, VI-4-56, numerous nymphs and adults.

Frankliniella insularis (Franklin)

Grain sorghum: Avondale, X-23-53 (Bill Zimmer); according to Watson (1923) infests many crops elsewhere.

Frankliniella minuta (Moulton)

Baileya multiradiata blooms: Pinal County, II-2-54; Apache Junction, IV-10-55. Sacaton, V-5-54.
Barley: Avondale, IV-14-55.
Brussels sprouts: Picacho, IX-14-53 (Jack Warren).
Encelia sp. prob. *farinosa:* Tempe, IV-9-55.
Oenothera sp. blooms: Litchfield Park, III-27-55.
Psilostrophe Cooperi blooms: Pinal Co., XI-7-54.

Frankliniella occidentalis (Pergande)

Alfalfa: Avondale, XI-2-53, damaging young seedlings (Bill Zimmer); El Mirage, I-6-58, numerous nymphs and adults; Gilbert, X-12-53; Perryville, III-30-55; Phoenix, II-3-54 (L. A. Hammers); I-6-58 (Stu

[1] Accepted for publication April 14, 1958.
Partial cost of publication of this paper was met by Arizona Fertilizers, Inc., Phoenix, Arizona.
[2] Indentification of thrips by either Kellie O'Neill or S. F. Bailey. Hosts plants about which there was doubt were identified by E. R. Blakley.
[3] With one record from California.

Evans); Pinal Co., X-19-53 (Dick Daley); Yuma, II-11-54 (Don McCain).
Baccharis sp. blooms: Phoenix, X-13-53 (Ben Fehrman).
Baileya multiradiata blooms: Pinal Co., II-2-54 and IV-10-55.
Brussels sprouts: Picacho, X-14-53 (Jack Warren).
Cabbage: Scottsdale, X-28-55.
Capsella Bursa-pastoris: Luke Field, II-24-57.
Castor bean: Phoenix, X-10-53.
Cauliflower: Peoria, XI-24-53 (L. A. Hammers).
Cercidium floridum blooms: Gila Bend, V-1-55.
Chenopodium sp.: Yuma, XII-1-53.
Cotton: Elfrida, VI-2-54; Eloy, VIII-14-57, from terminals (Skip Hornney); Marinette, VII-26-57, suspected of blasting flower buds (Bill Zimmer); Phoenix, VII-30-57, numerous in blooms and terminals (Bill Zimmer); Pinal Co., IV-30- and V-15-54.
Datura sp. blooms: Sacaton, VI-4-54.
Grape: Maricopa Co., V-5-54 (Joe Hartsig).
Gossypium Thurberi blooms: Sabino Canyon, IX-16-57.
Helianthus sp.: Phoenix, III-31-54.
Heterotheca subaxillaris blooms: Phoenix, X-13-53 (Ben Fehrman).
Johnson grass: Goodyear, XI-19-57 (Dale Berry); Phoenix, X-13-53 (Ben Fehrman).
Lettuce: Cochise Co., XI-10-53; Peoria, III-5-54; Phoenix, II-3-54 (L. A. Hammers); II-18-54; IX-23-54; II-7-55, numerous and probably damaging young seedlings in all fields (Bill Ritchie); Yuma, X-6-53; X-14-53, a pest of seedlings immediately after thinning and weeding (Don McCain); XI-30-53 (H. Czajowski).
Lupinus sp.: Tom Mix Monument, III-26-56.
Malva parviflora: Peoria, III-10-55.
Melon: Phoenix, VII-1-54.
Mesquite: Eloy, IV-28-54.
Mullein: Camp Wood, X-31-54.
Mustard: Pinal Co., X-19-53.
Nicotiana glauca: Litchfield Park, X-24-54.
Oenothera sp.: Litchfield Park, III-27-55.
Onion: Cochise Co., XI-23-53, from culls (C. L. Allen); Phoenix, II-18-54 (L. A. Hammers); III-11-54 (Bill Ritchie).
Pea foliage: Maricopa, I-9-58 (Ash and Nielsen).
Populus Fremontii galls: Pinal Co., IV-22-55.
Potato: Queen Creek, IV-23-54 (Ken Tipling).
Psilostrophe Cooperi blooms: Pinal Co., XI-7-54.
Sambucus sp.: Phoenix, V-26-55.
Sphaeralcea sp.: Maricopa Co., March and April, 1954.
Sida hederaceae: Yuma, IV-20-55.
Sisymbrium irio: Mesa, III-1-54; Phoenix, II-3-54.
Sugar beet foliage: Litchfield Park, II-1-58.
Sweetpea blooms: Avondale, IV-12-55.
Tamarix gallica: Maricopa Co., XI-24-53; Tempe, May, Safford and Tolleson, July; nymphs and adults common in flowers (Hopkins and Carruth 1954).
Yucca brevifolia: Tempe III-8-55 (E. R. Blakley).

Frankliniella tritici (Fitch), flower thrips

Baileya multiradiata: Sacaton, V-4-54.
Datura sp.: Sacaton, VI-4-54.
Psilostrophe Cooperi: Pinal Co., XI-7-54.

Haplothrips fasciculatus (Crawford)

Atriplex gall: Avondale, V-3-54.

Haplothrips purpuratus Hood or near

Atriplex sp.: Sacaton, VI-4-54.

Hercothrips marginipennis (Hood)

Corn: Maricopa, V-15-54, injury severe to bottom leaves (Herb Brown).

Heterothrips prosopidis Crawford

Mesquite: Eloy, IV-28-54.

Kurtomathrips morrilli Moulton

Bean: Salt River Valley (McKinney 1939).
Cotton: Gila Bend, severe injury (Bailey 1957).

Leptothrips sp. nr. *mali* (Fitch)

Aplopappus sp.: Tempe, II-26-54.

Microcephalothrips abdominalis (Crawford)

Baileya multiradiata blooms: Apache Junction, II-2-54; Pinal Co., IV-10-55.

Psilothrips paradalotus Hood

Atriplex sp.: Avondale, II-21-54.

Psilothrips priesneri (Moulton)

Lycium sp.: Casa Grande, IV-1-58, adults and a nymph.

Scirtothrips citri (Moulton), citrus thrips

Grapefruit foliage: Luke Field, XI-19-53 (L. A. Hammers).

Scirtothrips sp. (probably new)

Cotton: Mesa, VII-29-57, causing peculiar crinkling (not cupping) of leaves (R. L. Dezember); Phoenix, XI-19-53, numerous in one field (Ben Fehrman); Queen Creek, IX-28-56, terminal foliage peculiarly crinkled.
Other records on Cotton: Bard, California, VIII-27-54, leaves crinkled and plants bare of fruit (H. Czajowski); Mississippi, 1954 and Texas, 1953 and 1954, routine collection, no mention of injury.[4]

Scolothrips sexmaculatus (Pergande)

Cantaloupe: Phoenix, VI-17-54.
Robinia neomexicana: Douglas, IV-25-53 (C. L. Allen).

Thrips tabaci (Lindeman), onion thrips

Cauliflower: Peoria, XI-24-53.
Cotton: Maricopa Co., May, 1954; Pinal Co., IV-30-54; Scottsdale, V-5-54.
Lettuce: Peoria, III-5-54; Phoenix, II-18-54 (L. A. Hammers); III-5-53 (Bill Ritchie).
Onion: Cochise Co., XI-23-53, from culls; Phoenix, II-18-54 (L. A. Hammers); III-11-54 (Bill Ritchie); Tolleson, II-18-54.
Potato: Queen Creek, IV-23-54.
Sisymbrium irio: Phoenix, XI-3-54.

[4] Unpublished records of U. S. Dept. of Agriculture

REFERENCES CITED

Bailey, Stanley F. 1957. The thrips of California. Bull. California Insect Survey. 4(5): 143–220.

Hopkins, Lemac. and L. A. Carruth. 1954. Insects associated with salt cedar in Southern Arizona. Jour. Econ. Ent. 47(6): 1126–9.

McKinney, K. B. 1939. Common insects attacking sugar beets and vegetable crops of the Salt River Valley of Arizona. Jour. Econ. Ent. 32(6): 808–10.

Roney, J. N. 1949. Bermuda grass seed insects in Arizona. Jour. Econ. Ent. 42(3): 555.

Watson, J. R. 1923. Synopsis and catalog of the Thysanoptera of North America. Univ. Florida Agric. Expt. Sta. Bull. 168, 100 pp.

Malathion Resistance Studies on the House Fly[1]

R. W. FAY, JOHN W. KILPATRICK, and G. CLINTON MORRIS, III

ABSTRACT

Malathion resistance in house fly (*Musca domestica* L.) strains from Phoenix, Arizona, and Savannah, Georgia, was confirmed in laboratory tests during the fall of 1956. Physiological resistance was shown using exposures to malathion-sugar residual applications and topical applications of malathion-acetone solutions. Using colony cages with 5, 25, and 45% of the available resting surface treated with malathion-sugar residual deposits, strains with higher levels of resistance were selected from the resistant field strain. The physiologically resistant strains also showed a correlated behavioristic resistance in their avoidance of malathion baits but not of Dipterex baits. Comparison of resistance levels of 1-, 3-, and 5-day-old adult males and females showed a marked inverse relationship of age to resistance.

During the fall of 1956, two strains of the house fly (*Musca domestica* L.) were tested in the laboratory for resistance since residual deposits and poison baits of malathion were failing in field applications. The first strain was taken at a dairy in Phoenix, Arizona, the second from the Bethesda dairy near Savannah, Georgia (Kilpatrick & Schoof 1957). The present paper deals with preliminary evaluations on both strains and with studies on strains derived from the Bethesda field stock through further laboratory selection.

In initial tests, adult flies of a standard insectary strain (NAIDM[2]-1948) and of the suspect strains (Phoenix-field and Bethesda-field) were exposed to residual deposits of 200 mg. of malathion-500 mg. of sugar per sq. ft. An emulsion spray was formulated from a 25% malathion-xylene concentrate containing 2% Triton X-155 by mixing 1:4 with a sugar-water solution to give a finished emulsion of 5% malathion-12.5% sugar. This emulsion was applied to plywood panels at the rate of 4 ml. per sq. ft. to prepare the test residues. For additional comparison of the three strains, tests were made with deposits of 200 mg. of DDT per sq. ft. from a xylene emulsion spray. Results (table 1) showed the Phoenix-field strain to be less malathion-resistant than the Bethesda-field strain and further studies were continued with the latter.

The Bethesda-field strain was placed under laboratory selection by establishing colonies in cages with 5, 25, or 45% of the available resting surface treated with residual deposits of 200 mg. of malathion-500 mg. of sugar per sq. ft. Insectary-reared pupae of the Bethesda-field strain were placed in each colony and the emerging adults were supplied with food and water. Eggs were collected from surviving adults after the fifth day and reared in the insectary as generation F_1. The strains were designated as Bethesda-5, Bethesda-25, and Bethesda-45 according to the degree of colony-cage treatment. The parent generation in the Bethesda-5 colony had large numbers of surviving adults and a good supply of eggs. The Bethesda-25 colony had fair numbers of surviving adults but only enough eggs to restock the F_1 generation pupae. The Bethesda-45 colony had few surviving adults and no eggs. F_1 pupae for the Bethesda-45 colony were secured from eggs of the Bethesda-5 adults. By the F_2 generation, enough eggs were secured from surviving adults of the Bethesda-45 strain to restock the next generation.

In the F_4 generation, adults of the three laboratory-selected strains were compared with the wild Bethesda-field strain (new field collections) and the NAIDM-1948 flies using topical applications of technical malathion in acetone. The applications were made to the ventral surface of the thorax of the 3-day-old adult females using a microsyringe delivering drops of 0.5 mm.[3]. The micrograms of malathion required for mortality levels of 10, 50, and 90% for the various strains are shown in table 2.

Although the median lethal dose of the Bethesda-field strain was only two-fold over that of the susceptible strain, difference at the 90% mortality level was very marked. The laboratory Bethesda-5 strain was apparently under approximately the same degree of selection pressure as the wild Bethesda-field strain. The further increases in the LD-50 and LD-90 values for the Bethesda-25 and Bethesda-45 strains indicated that the wild strain

[1] From the Communicable Disease Center, Bureau of State Services, Public Health Service, U. S. Department of Health, Education, and Welfare, Savannah, Georgia. Accepted for publication January 16, 1958.

[2] National Association of Insecticide and Disinfectant Manufacturers.

Table 1.—Twenty-four hour mortality (%) of adult house flies of three strains when exposed for 30-minute periods to residual deposits of 200 mg. of malathion-500 mg. of sugar per sq. ft. and of 200 mg. of DDT per sq. ft.

STRAIN	MALATHION-SUGAR		DDT	
	Males	Females	Males	Females
NAIDM-1948	94	62	99	96
Phoenix-field	78	38	64	55
Bethesda-field	13	6	3	2

Table 2.—LD-10, LD-50, and LD-90 values for topical application of malathion-acetone solutions applied to the ventral surface of the thorax of 3-day-old adult female house flies of five strains.

Strain	Micrograms of Malathion per Fly		
	LD-10	LD-50	LD-90
NAIDM-1948	0.35	0.48	0.56
Bethesda-field	0.49	1.17	4.40
Bethesda-5 (F_4)	0.53	1.50	4.00
Bethesda-25 (F_4)	1.60	5.00	16.00
Bethesda-45 (F_4)	2.40	6.60	24.00

of field flies did not at that time reflect the full extent of possible resistance to malathion.

Field observations showed that flies of the Bethesda-field strain avoided malathion baits but were attracted to Dipterex or DDVP baits (Kilpatrick & Schoof 1957). Laboratory measurements were made of this phenomenon using 3-day-old adult females of the susceptible, the field, and the three laboratory-selected strains. Adult flies were allowed to feed for a 30-minute period and were then anesthetized with CO_2 to permit the selection of 200 adult females. These flies appeared to be fully recovered from any effects of the carbon dioxide anesthesia within 2 hours. After a 4-hour starvation period, the adult females of each strain were placed in separate 18×16×14-inch wire-screen test cages. The test bait, confined in a ½-inch length of 2-inch diameter tubing, was centered on the floor of each cage. Flies on the bait were counted at 10-minute intervals and the knocked-down and dead flies were recorded at the end of 1 hour. The test bait was removed and the insects were supplied with food and water to determine the subsequent 24-hour mortality. Results of these tests (table 3) confirmed the lower numbers of flies of the Bethesda-field strain observed on the malathion bait. Further decrease in the number of flies on the bait occurred as the malathion resistance levels were increased in the B-25 and B-45 selected strains. Comparable tests using Dipterex baits indicated little or no difference in their attraction for or effectiveness against the susceptible and the malathion-resistant strains. Results with the malathion bait based on the 1-hour and the 24-hour mortality values (table 3) agreed quite closely with the residual and topical evaluations of resistance.

The reactions of individual flies of the various strains to the malathion baits were observed. The susceptible flies landed on the bait or close to it and upon contact moved over the material feeding for a period of several minutes. The resistant flies, however, were never seen to land directly on the bait and those which landed near it approached but upon the first contact flew off in a matter of one or two seconds.

During April and May, 1957, flies were collected again at the Bethesda dairy and compared with the NAIDM-1948 and the B-45 strain to determine possible changes in resistance levels during the intervening months. Using 30-minute exposures to residual deposits of 200 mg. of malathion-500 mg. of sugar per sq. ft., 24-hour mortalities for males and females of the NAIDM-1948 strain were 85 and 66%, respectively. Similar tests with the Bethesda-field strain gave 74 and 41% mortalities, compared with

Table 3.—Results with 1-hour exposures of 200 3-day-old adult female house flies of five strains to 2% malathion or 1% Dipterex dry baits.

Strain	Average Number on Bait	Per Cent Mortality at	
		1 Hr.	24 Hr.
	Malathion dry bait		
NAIDM-1948	5.0	59	69
Bethesda-field	2.0	21	25
Bethesda-5	4.0	32	37
Bethesda-25	0.8	3	6
Bethesda-45	0.5	0	2
	Dipterex dry bait		
NAIDM-1948	18.0	66	91
Bethesda-field	14.0	48	77
Bethesda-5	19.0	86	97
Bethesda-25	19.0	53	92
Bethesda-45	19.0	81	95

values of 13 and 6% obtained with adults collected in the fall of 1956. With the Bethesda-45 F_{11} generation, the mortalities were 1 and 0%. Further tests using topical applications of malathion confirmed that some loss of resistance had occurred over the winter in the Bethesda-field strain. The B-45 strain, however, showed greater homogeneity for resistance, with considerable reduction in the more susceptible portions of the strain.

In preparation for further resistance selection, the effectiveness of topical applications of malathion was determined using 1-, 3-, and 5-day-old adult males and females of the F_{14} generation of the B-45 strain. The data (table 4) show the 1-day-old males and females to be considerably more resistant than 3- or 5-day-old adults to topical applications of malathion.

Although the appearance of malathion-resistant strains in the field have been limited to a relatively few locations, the evidence of a combined physiological and behavioristic-resistance pattern in the present studies portend the possible future relegation of malathion to those insecticides which have become ineffective against the house fly.

Table 4.—LD-10, LD-50, and LD-90 values for topical application of malathion-acetone solutions applied to the ventral surface of the thorax of 1-, 3-, and 5-day-old adults of the B-45 strain of malathion-resistant house flies (F_{14} generation).

Sex	Age (days)	Micrograms of Malathion per Fly		
		LD-10	LD-50	LD-90
Male	1	5.4	9.0	15.0
	3	1.8	3.5	6.0
	5	1.0	1.8	3.0
Female	1	8.8	18.0	38.0
	3	4.6	9.2	20.0
	5	2.7	5.5	11.0

References Cited

Kilpatrick, John W., and H. F. Schoof, 1957. A field strain of malathion-resistant house flies. Jour. Econ. Ent. 51(1): 18–19.

Laboratory and Field Tests with the Granulosis of the Red-Banded Leaf Roller[1]

E. H. Glass, *New York State Agric. Expt. Sta., Geneva*

Abstract

A granulosis caused by a virus, *Bergoldia clistorhabdion* (W. and S.) has been observed in larvae of the red-banded leaf roller, *Argyrotaenia velutinana* (Wlk.) in an occasional apple orchard in New York. The disease has not been observed to be abundant enough to influence appreciably population levels. In the laboratory first instar larvae readily contracted the disease when fed on apple leaves treated with suspensions of triturated diseased larvae at dilutions from 1 last instar larva per 1000 ml. of water to 1 larva per 512,000 ml. of water. Disease symptoms and death developed more rapidly at the higher concentrations than at the lower. Suspensions of triturated diseased larvae at rates of 5, 50 and 100 larvae per 100 gallons sprayed on apple trees during the hatching of first brood eggs infected and destroyed most larvae, but not before they damaged many fruits.

Until DDT was introduced into the apple spray program in 1946, the red-banded leaf roller, *Argyrotaenia velutinana* (Wlk.), was considered a minor pest of apples even though it had occasionally caused localized damage in outbreak years. Over the past 10 seasons it has become a major pest on apples but even during this period there have been large population fluctuations from year to year and from orchard to orchard. These variations have raised the question of the influence of biological control agencies.

During the past nine seasons the writer has made field observations on the leaf roller in many commercial apple orchards. There have been no indications that predators or parasites have been a limiting factor at any time. Observations made on large numbers of eggs and larvae collected for laboratory and insectary work have disclosed only minor biological control activity, certainly not enough to have greatly influenced population trends in commercial plantings. Several writers have ascribed this low level of parasites and predators to the extensive use of long-residual contact insecticides such as DDT.

In 1949 Sibold (1950) discovered a granulosis in insectary reared red-banded leaf roller larvae. The virus causing this disease was named *Bergoldia clistorhabdion* by Wasser & Steinhaus (1951). Schoene & Sibold (1952) reported that the disease was readily found in all of a number of Virginia orchards surveyed in October of 1950. They believed granulosis had greatly reduced the leaf roller population in that area. The disease has been observed to be widespread in northern Virginia and in southern Indiana by C. H. Hill and D. W. Hamilton, respectively (private communications).

Granulosis and its symptoms in red-banded leaf roller have been described by Wasser & Steinhaus (1951), Sibold (1950) and Schoene & Sibold (1952). This paper is a report of field observations and of laboratory and field tests with this disease in New York.

Field Observations.—In September 1950 the writer collected approximately 100 nearly mature red-banded leaf roller larvae from an orchard in Geneva and sent them to Professor W. J. Schoene in Virginia. He reported 65% were diseased. Very few red-banded leaf roller have appeared in this orchard in spite of the fact that very little spraying has been done since that time. However, no disease symptoms have been observed in those larvae found during the intervening period so it is questionable that the low population level can be attributable to granulosis.

Elsewhere in the state, the writer has observed very few larvae showing disease symptoms. In 1953, larvae were collected from nine orchards and reared in the insectary to the adult stage. Granulosis was present in a collection from only one of these plantings and even here it affected only 28% of the individuals. It is of interest to note that survival was 87%, indicating a very low level of parasitism. A small number of granulosis infected larvae were observed in two orchards in 1956. Otherwise none have been seen. The number of diseased individuals seemed to be too few to effect much control of the leaf roller in New York.

Laboratory Tests.—The method adopted for handling leaf roller larvae under laboratory conditions was to rear them individually in 9×2.5 mm. shell vials closed with cork stoppers. This was done to prevent unintentional initiation of the disease and to localize its spread. The apple leaves used as food kept in good condition for 1 to 2 weeks in these vials. Usually a fresh leaf or two had to be added to insure an adequate food supply for completion of larval development at 80° F. Considerable care had to be exercised to avoid contamination during this operation.

The technique for innoculating larvae was to triturate a diseased larva in a mortar in a small amount of tap water. This suspension was then diluted to the desired concentration. Last instar larvae in the advanced stages of the disease were selected for this work. No attempt was made to determine the concentration of virus particles in the suspensions used and dosages are given as the number of diseased larvae per unit of water. Enough B-1956 was added to insure wetting of apple leaves dipped into the suspensions. The leaves were placed on paper towels until dry and then put into the shell vials. As a general practice all operations proceeded from the lowest to the highest concentrations in a series. Water checks handled in the same manner were usually set up at the beginning and end of each series to determine the amount of contamination.

Newly hatched larvae were then placed on the leaves and the vials stoppered. Most of the larvae used were from field collected egg masses which were held in shell vials until hatched and then transferred to the leaves with a camel-hair brush. Larvae tend to spin down on a strand of silk and thus it was possible to avoid touching treated leaves with the brush. Vials were kept in racks in an 80° F. constant temperature room. The vials were not opened except to add a fresh leaf when needed. This procedure prevented close examination of larvae to deter-

[1] Journal Paper No. 1105, New York State Agricultural Experiment Station, Geneva. January 10, 1958. Accepted for publication January 16, 1958.

Table 1.—Concentration of granulosis suspensions in relation to disease incidence in the laboratory, 1954.

TREATMENT (ML. WATER PER 1 LARVA)	DISEASE AT 13 TO 15 DAYS			PER CENT PUPATING		PER CENT MOTHS	
	May 10 Series[a]	May 26 Series[b]	June 4 Series[b]	May 26 Series	June 4 Series	May 26 Series	June 4 Series
1,000	80						
2,000	63	most		0		0	
4,000	80						
8,000	85	most	all	3	15	0	0
16,000	78						
32,000		most	all	3	20	0	8
128,000		most	all	28	35	0	3
512,000		most	most	35	44	3	18
Check (before handling diseased material)	8	0	none	51	78	42	63
Check (after handling diseased material)	53	few		75		50	

[a] After examination, diseased larvae removed for field tests.
[b] Vials not opened, based on observations of larvae visible through glass.

mine the exact percentage of disease and thus only general observations were made on the presence or absence of the disease. Survival to the pupal and adult stages were judged the best criteria for determining the extent and intensity of the disease.

The source of all disease material used in this study was that collected June 1, 1953 from the JiMeva farm. The supply was enlarged from time to time in the laboratory as needed for field testing. Diseased larvae were kept in the freezing compartment of a household refrigerator for later use. Results indicated little or no loss of virulence from such storage over a 2-year period.

Preliminary tests showed that a concentration of 1 larva per 20 ml. of water was very infectious to newly hatched larvae. In one test, 60% were dead within a week after exposure and most of the remainder were smaller than those in the check. Twenty-four per cent reached the pupal stage but no moths emerged. Many of the check larvae became diseased in this test but most developed to the pupal and moth stages. On the basis of such tests, dilution series were conducted to determine what concentrations might be needed to effect field control. The results of these tests are shown in table 1.

The May 10 series was sacrificed after 15 days for use in field tests. Examination at that time showed there was no reduction in the incidence of granulosis as the dilution rate was increased from 1 larva to 1000 ml. to 1 larva in 16,000 ml. The check series set up following the treatments became 53% diseased. However, the disease was not as advanced in these checks as in the innoculated groups. In the May 26 and June 4 series, the disease was present even at the concentration of 1 larva per 512,000 ml. (approximately 1 larva in 135 gallons). However, at this low concentration more larvae pupated and a few reached the moth stage. In some preliminary tests made in 1953 a few individuals reached the adult stage even at the dilution of 1 larva per 2000 ml.

These laboratory studies show that with the technique used the granulosis can be spread at very low dilutions of the innoculum but that the speed of disease development and death is correlated with the dilution. Mortality may occur in the first instar at high levels and not at all or until the pupal stage at low concentrations. No work was done to determine whether moths developing from diseased larvae could oviposit normally or what the condition of such eggs might be if they were laid.

In the eastern apple growing regions growers normally apply fungicides in ten or more sprays each season. The materials most commonly used are elemental sulfur, ferbam, captan, glyodin and organic mercury compounds. Preliminary tests had shown that none of the fungicides were toxic to the larvae at the concentrations used. However, if such materials inhibit the granulosis or prevent it from spreading they could limit the usefulness of the disease in leaf roller control. To obtain some information on this point, granulosis suspensions prepared as described in the foregoing were mixed with some of the fungicides at concentrations normally used in the field. Leaves were dipped in these mixtures and larvae added as described earlier. Results are given in table 2.

The results of the May 10, 1954 series show that none of the fungicides reduced the incidence of disease with the possible exception of glyodin. The May 27 series was made with a lower concentration of the virus. Observation showed the disease to have been present in all the series except the glyodin and the check. Many larvae pupated

Table 2.—Effects of fungicides on the incidence of granulosis in laboratory tests. 1954.

TREATMENT	PER CENT DISEASED 13 TO 15 DAYS	PER CENT PUPATED	PER CENT MOTHS
May 10 Series[a]			
1 larva per 1,000 ml. water	80	—	—
Same+Tag[b] 0.5 pt.[c]	75	—	—
Same+sulfur 5 lbs.	95	—	—
Same+50% captan 2 lbs.	80	—	—
Same+76% ferbam 1.5 lbs.	88	—	—
Same+dichlone 0.5 lb.	78	—	—
Same+34% glyodin 2 qts.	50	—	—
Water check	8	—	—
May 27 Series			
1 larva per 128,000 ml. water	some	43	5
Same+Tag 0.5 pt.[a]	some	56	0
Same+sulfur 5 lbs.	some	25	3
Same+50% captan 2 lbs.	some	49	5
Same+76% ferbam 1.5 lbs.	some	49	5
Same+dichlone 0.5 lb.	some	55	0
Same+34% glyodin 1 qt.	none	65	57
Water check	none	60	49

[a] Examined for disease and sacrificed for field test inoculum after 15 days.
[b] Phenylmercury acetate.
[c] Fungicides rate per 100 gallons.

Table 3.—Control of first brood red-banded leaf roller with granulosis sprays in the L. Salisbury Orchard, 1954.

Plot No.	Material and Quantity per 100 Gallons	Applied: Petal Fall[a]	Applied: 10-Day[b]	Per Cent Fruit Injury
1	50% TDE, 1 lb.		x	0.0
2	15% parathion, 1 lb.		x	0.4
3	100 diseased larvae	x		27.7
4	50 diseased larvae		x	40.5
5	5 diseased larvae		x	28.5
6	Check			54.0

[a] May 21—No eggs hatched, first hatching occurred May 22.
[b] June 3—97% of eggs hatched.

in all treatments but moths developed in numbers only in the glyodin and check series. It would appear that only glyodin reduces the incidence of the disease.

FIELD TEST.—Control experiments have been conducted over a period of years to determine satisfactory chemical control measures for the red-banded leaf roller. Details of the technique used are given by Glass & Chapman (1952). Briefly the method involves thorough spraying of apple trees in heavily infested orchards. Results are measured by scoring the larval feeding scars on the fruits. In 1954, three granulosis treatments were included in a test of a number of insecticides against the first brood. Diseased larvae were triturated in a small amount of water in the same manner as for the laboratory work. The concentrate suspensions were added to the spray tank in the field and applied to the trees as sprays. All treatments except 4 and 5 were replicated on four single tree plots in a randomized block design. Plots 4 and 5 were applied only to single trees. The results are given in table 3. Severe fruit damage in the check plot shows that there was a very heavy infestation present in the orchard. Of the several insecticides tested, only results with two standard control agents are given. It is obvious that the granulosis treatments were not effective in preventing fruit injury as compared with TDE and parathion. The apparently inconsistent results in plots 4 and 5 were probably the result of natural variation and lack of replication. In addition to the modest reduction of the percentage of fruit injury in the granulosis plots, there was also a reduction of the extent of damage on injured fruits. This was more evident on plot 3 then on plots 4 and 5 which were treated 13 days later. The disease inactivated the larvae before they had completed development but not before some fruit feeding occurred.

On June 18 forty larvae were collected from a check tree and 19 from Plot 5 and placed in shell vials for observation in the laboratory. In the check 98% reached the pupal stage, 53% the moth stage. In the Plot 5 group only 50% pupated and none survived to the moth stage.

Samples of the spray suspensions used in the field were brought back for laboratory evaluation. The results given in table 4 show that all three concentrations were highly virulent and ultimately lethal. It is evident that granulosis can be easily spread in the field but that it does not inactivate larvae soon enough to prevent fruit damage during the current season and thus cannot be substituted for chemical control of severe infestations.

DISCUSSION.—The foregoing report can be considered a progress report on a study initiated to determine the possible use of granulosis for the control of red-banded leaf roller. In most cases the work has gone only far enough to suggest and outline areas of further study and testing.

Table 4.—Laboratory results on first instar red-banded leaf roller with diseased larval suspensions used in the Salisbury Orchard, 1954.

Treatment	Observed Granulosis in Larvae	Per Cent: Pupated	Per Cent: Moths
May 26 Series			
Water check	none	60	49
Plot 3 (100 larva/100 gal.)	most	0	0
June 4 Series			
Water check	none	78	63
Plot 4 (50 larvae/100 gal.)	all	5	0
Plot 5 (5 larvae/100 gal.)	all	28	3

The practical employment of a virus or microbial organism to control insect pests of apples could be somewhat limited by the extensive use that growers make of certain fungicides. The granulosis virus in question does not appear to be affected in the laboratory by the commonly used apple fungicides (with the possible exception of glyodin). The disease was not noticeably inhibited by wettable sulfur sprays used for apple scab control in the field test in 1954.

Another consideration is the time required for the disease to inactivate or kill the leaf roller larvae. In the present case a disease must prevent fruit injury if it is to be used as a direct substitute for chemical control. The results obtained both in the field and laboratory suggest that granulosis kills too slowly to prevent such injury unless very high concentrations are used. Such high levels would likely be prohibitively expensive. Therefore, it appears that the practical use of granulosis could be to reduce the population level and thus indirectly to keep fruit damage to minimum levels in subsequent years.

The question of how long a general application of a granulosis spray would be effective in suppressing the leaf roller is not known. The writer believes the innoculum of such a treatment would have to remain effective at least 2 years to make an application worthwhile in view of the probable cost of the treatment and the occurrence of reinfestation from outlying areas.

Another problem is to find means of producing the virus easily and cheaply. The leaf roller can be successfully reared in the laboratory or greenhouse but mass rearing techniques have not been developed. The success of efforts to mass rear this insect with minimum labor will have a great bearing on the practical use of granulosis. Culture of virus in a medium other than the leaf roller larva is of course a possibility. Steinhaus (1951) has reported on limited unsuccessful attempts to propagate two other granulosis viruses in chick embryos. In any event the development of some reasonable method of producing granulosis innoculum in quantity at a reasonable cost is essential to any wide scale use of the disease in orchard control of the red-banded leaf roller.

REFERENCES CITED

Glass, E. H., and P. J. Chapman. 1952. Red-banded leaf roller and its control. New York State Agric. Expt. Sta. Bull. 755.

Sibold, N. V. 1950. Granulosis of the red-banded leaf roller. Virginia Jour. Sci. 1(3): 226.
Schoene, W. J., and N. V. Sibold. 1952. A virus disease of the red-banded leaf roller. Jour. Econ. Ent. 45(6): 1081.
Steinhaus, E. A. 1951. Report on diagnosis of diseased insects 1944–1950. Hilgardia. 20(22): 629–78.
Wasser, H. B., and E. A. Steinhaus. 1951. Isolation of a virus causing granulosis in the red-banded leaf roller. Virginia Jour. Sci. 2(2): 91–93.

The Effect of Spotted Alfalfa Aphid Protection by Systemic Insecticides and Varieties on Alfalfa Production the Year Following Seedling Protection[1]

R. C. DOBSON[2]

ABSTRACT

Yields of six different varieties of alfalfa, each treated at seeding time with granulated Thimet, Bayer 19639 and Bayer 23129 were studied to determine the effects of an infestation by the spotted alfalfa aphid (*Therioaphis maculata* (Buckton)) in the seedling stage on yield of variety and treatment. The two tolerant varieties, Lahontan and New Mexico 16 were not seriously injured by the aphid attack during the seedling stage. Ranger and African, classed as semi-tolerant, were injured but recovered so that at the end of the first production year they produced as much alfalfa as the best treated plots.

Bayer 19639 applied as a granulated material at seeding time protected non-tolerant varieties from aphid attack in the seedling stage and the first production year's yield was as great as protected tolerant varieties.

The spotted alfalfa aphid (*Therioaphis maculata* (Buckton)) has been the primary pest of alfalfa in New Mexico since its invasion of the state in 1953. It was responsible for a 4 million dollar loss in production in 1954 and since that time has attacked alfalfa in 30 states (Staten 1954).

Protecting seedling stands of alfalfa from this insect has become a necessity in New Mexico. Effects of systemic insecticides on stand and growth of several varieties of seedling alfalfa have been reported by Dobson & Watts (1957). Production figures have not been previously reported showing effects on yields of mature alfalfa due to spotted alfalfa aphid attacks on seedling stands. The work reported here shows the effect of this pest on yield of several varieties for the production year following fall attack on seedling alfalfa. It shows also the difference in production between stands of several varieties of alfalfa protected during the seedling stage and those which were unprotected.

PROCEDURES.—Six alfalfa varieties and four treatments were tested in a split plot design as reported by Dobson & Watts (1957). The varieties were replicated six times and each 40-foot variety plot was subdivided into 10-foot sections to accommodate the four treatments.

A heavy population of aphids built up on the seedling stands in the fall of 1956 as reported previously (Dobson & Watts 1957). During 1957, economic infestations of this aphid failed to materialize in this area. Insecticidal treatments were not applied to the experimental plots during 1957.

The varieties used in this experiment were classed in the following manner: New Mexico Common and New Mexico 11-1 as susceptible, Ranger and African as semi-tolerant, and New Mexico 16-1 and Lahontan as tolerant to this aphid.

Table 1.—Green weights of first-year cuttings of six varieties of alfalfa treated with insecticides for control of seedling infestation by the spotted alfalfa aphid.[a]

VARIETY	GREEN WEIGHTS (LBS.)					
	Bayer					
	Thimet	19639	23129	Untreated Check	Total[b]	Average[c]
New Mexico Common	37.6[d]	161.7	91.4	7.1	297.8	12.41
New Mexico 11-1	72.8	180.8	134.6	13.8	402.0	16.75*
Ranger	101.2	158.1	136.2	86.2	481.7	20.07**
African	110.4	172.7	150.4	47.2	480.7	20.03**
Lahontan	133.1	154.7	145.5	138.9	572.2	23.89**
New Mexico 16	152.2	178.9	151.8	143.2	626.1	26.09**
Total	607.3	1006.9	809.9	434.4		
Average	16.81**	27.97**	22.5**	12.122		

[a] Insecticides applied at planting time, September 27, 1956.
[b] From 388 feet of row.
[c] From 32 feet of row.
[d] From 48 feet of row.
L.S.D. for treatments, at 5% = 1.541, at 1% = 2.131; for varieties, at 5% = 3.797, at 1% = 5.955.
* Significant at 5% level.
** At 1% level.

The insecticides used, all classed as systemics, were Thimet, Bayer 23129 (S-2-(ethylthio)ethyl *O,O* dimethyl ester phosphorodithioic acid), and Bayer 19639 (also called Disyston). As has been reported, 1 pound actual per acre (granulated) of the materials was applied to the seed furrow at planting time. Readings on stand and plant growth throughout the fall of 1956 have been reported by Dobson & Watts (1957). Yield data for 1957 were taken by hand cutting the plots six times during the season (table 1). The alfalfa was allowed to come into about one-tenth bloom before each cutting. All yield data are calculated on a green weight basis. The weights of alfalfa were taken only from the center 8 feet of each row, leaving a one-foot buffer at each end of the plot.

RESULTS.—Analysis of the figures on total production for the season showed that all varieties tested produced significantly more alfalfa than the check variety, New Mexico Common. The amount of green weight alfalfa

[1] Journal Series No. 118, Agricultural Experiment Station, New Mexico College of Agriculture and Mechanic Arts, State College. Accepted for publication January 14, 1958.

[2] Assistant Entomologist, Department of Botany and Entomology, New Mexico College of Agriculture and Mechanic Arts.

Table 2.—Yields from alfalfa variety plots treated with Bayer 19639.

Variety	Weight (lbs.)	
	Total[a]	Average[b]
Ranger	158.1	4.392
African	176.9	4.914
New Mexico Common	161.7	4.492
New Mexico 16	178.9	4.969
New Mexico 11-1	180.8	5.022
Lahontan	154.7	4.297
L.S.D. at 5% level		.583
at 1% level		.914

[a] From 288 feet of row.
[b] From 8 feet of row.

produced by New Mexico 11-1, an improved aphid susceptible variety, was significantly higher than the check at the 5% level. The other varieties, New Mexico 16, Lahontan, African, and Ranger, produced more than the check. These differences were significant at the 1% level (table 1). The total production analysis showed that all treatments, Thimet, Bayer 19639, and Bayer 23129, effectively increased yields of alfalfa over the untreated plots. These increases were significant at the 1% level (table 1).

When individual variety analyses were made, a distinct pattern emerged. The total year's productions of New Mexico 16 and Lahontan were increased significantly by treatment with Bayer 19639. These increases, while significant in the aggregate for the year, were not large enough to show up when individual harvest-weight analyses were made.

The year's total hay production of the semi-tolerant variety, African, was increased significantly by treatment with Bayer 19639 and Bayer 23129, but not by the Thimet treatment. This increase in production was great enough to show in individual harvest weights. The difference between the Bayer 19639 and untreated plots was great enough in each of the six cuttings to be significant at the 5% level. The yield from the plots treated with Bayer 23129 was greater than from the untreated plot through the first five cuttings, but not for the sixth cutting. This difference was significant at the 5% level.

Total production for the year of Ranger, the second semi-tolerant variety, was increased significantly by treatment with Bayer 19639 but not by treatment with Thimet or Bayer 23129. The increase in production by treatment with this material was significant at the 5% level through the fourth cutting, after which the untreated plots produced as well as the plots treated with Bayer 19639.

The non-tolerant variety, New Mexico Common, showed increases in total production for the year in the plots treated with Bayer 19639 when compared with the untreated plots. The analyses of each harvest showed that the treated plots produced more alfalfa at each cutting than the untreated plots. The increase was significant at the 5% level to all cuttings.

New Mexico 11-1, the other non-tolerant variety, showed total yearly production increases in both the plots treated with the Bayer 19639 and the Bayer 23129 when they were compared with untreated plots. The separate cutting analyses showed that the plots treated with Bayer 19639 and 23129 produced more alfalfa at each cutting than the untreated plots. The differences between 19639 and untreated check were significant at the 1% level but only at the 5% level when the Bayer 23129 and untreated check were compared.

When the plots of all the varieties treated with Bayer 19639 were compared with each other, no differences in production were found (table 2). Since production records, taken on the New Mexico College of Agriculture and Mechanic Arts Agronomy Farm, showed that New Mexico Common and New Mexico 11-1 routinely produced, in the absence of the spotted alfalfa aphid, as much if not more alfalfa per season than the other varieties tested, it is evident that some permanent damage to these stands was caused by the attack of the aphid in spite of the protection afforded them.

References Cited

Dobson, R. C., and J. G. Watts, 1957. Spotted alfalfa aphid occurrence on seedling alfalfa as influenced by systemic insecticides and varieties. Jour. Econ. Ent. **50**(2): 132–5.

Staten, G. 1954. Unpublished report to Director of New Mexico A & MA Experiment Station.

NOTICE TO AUTHORS

Each manuscript submitted for publication in the JOURNAL OF ECONOMIC ENTOMOLOGY must be accompanied by an abstract suitable for publication, and prepared in accordance with instructions for the preparation of abstracts as required by *Biological Abstracts*. This abstract will replace the usual author's summary. See "Important Notice to Authors" which appeared in the *Bulletin of the Entomological Society of America* 3(2): 1. June, 1957.

Granulated Insecticides for the Control of Seed Weevils on Crimson Clover[1]

H. H. Tippins, *Georgia Experiment Station, Experiment*

Abstract

Field experiments conducted at three locations in Georgia in 1956 and 1957 involved broadcast applications of granulated formulations of several insecticides for the control of *Hypera meles* (F.) and *Hypera nigrirostris* (F.). Results of the tests in 1956 showed that aldrin, dieldrin, or heptachlor reduced weevil populations significantly. Clover seed yields were consistently higher in plots receiving heptachlor or dieldrin. Tests in 1957 compared two application dates and several rates of application of dieldrin and heptachlor. The results showed that all rates of each insecticide controlled the weevils regardless of the date of application. Control of the weevils resulted in increased seed yields.

The clover weevils, *Hypera meles* (F.) and *Hypera nigrirostris* (F.), have been known as pests of red clover for many years (Detwiler 1923) in the eastern United States. Only recently they have been reported as damaging crimson clover seed crops in the South. Johnson & Nettles (1953) reported heavy infestations of both species in South Carolina. The weevils were reported from two counties in Georgia by Beckham (1956). Recent surveys in Georgia have revealed that the insects are present in 41 of 45 counties surveyed. They probably occur in all except the southernmost counties of Georgia. *Hypera meles* seemed to be the more important of the two from the standpoint of damage to crimson clover.

The principal damage to crimson clover by both of the insects is caused by the larvae feeding on the flowers, ovules, and growing seeds. Economic damage by the adults is caused mainly by their feeding on the stems, which results in lodging of the flower heads.

Both *Hypera meles* and *H. nigrirostris* are active from February until June. Oviposition begins during late March for *H. meles* and a week or so earlier for *H. nigrirostis*. Larval populations reach a peak about May 1. The adults spend the summer, fall, and early winter months in an inactive condition under protective cover in or near clover fields.

The period of reproductive growth of crimson clover coincides with the period of larval development of the two weevils. Thus, crimson clover is an ideal host plant. The general practice in the production of crimson clover seed is to allow livestock to graze until about the last week of March, at which time they are removed and the clover is allowed to grow and produce seed.

Beckham (1956) reported that granulated aldrin, dieldrin, or heptachlor at 2.5 pounds of the actual toxicant per acre gave outstanding control of *H. meles* and *H. nigrirostris*. He concluded that granulated insecticides should be applied about the third week of March. Hansen & Dorsey (1957) showed that granulated dieldrin or heptachlor at the rate of 0.5 pound of the active material per acre controlled *H. nigrirostris* but had little effect on *H. meles*.

Field experiments for the control of *Hypera meles* and *H. nigrirostris* on crimson clover were conducted in Georgia in 1956 and 1957. The results of these tests are reported in this paper.

Table 1.—Control of weevils on crimson clover with granulated insecticides. Fayette County and Putnam County, Georgia, 1956.

Treatment and Rate[a]	Adults per 10 Sweeps		Larvae per 10 Sweeps	Pounds of Seed per Acre
	H. meles	*H. nigrirostris*	(both species)	
	Fayette County, Test 1			
Heptachlor 2.5	2.25	0.25	0.00	1,114
Dieldrin 2.5	2.00	0.00	0.75	1,003
Aldrin 2.5	2.00	1.00	0.75	996
Toxaphene 10.0	6.00	1.50	7.00	559
Untreated	11.25	3.75	21.25	511
L.S.D. at 5% level	5.53	2.40	9.28	242
at 1% level	7.76	n.s.	11.41	339
	Fayette County, Test 2			
Heptachlor 2.5	2.00	0.50	0.00	856
Dieldrin 2.5	2.50	0.75	0.25	673
Aldrin 2.5	3.50	1.00	0.00	671
Toxaphene 10.0	11.00	2.75	3.25	442
Untreated	12.00	2.75	3.25	500
L.S.D. at 5% level	4.11	1.88	2.81	256
at 1% level	5.76	n.s.	n.s.	n.s.
	Putnam County, Test 3			
Heptachlor 2.5	1.50	0.50	0.25	700
Dieldrin 2.5	8.00	1.00	2.50	762
Aldrin 2.5	4.75	1.25	1.50	687
Toxaphene 10.0	10.50	2.00	12.75	601
Untreated	21.00	2.75	19.00	597
L.S.D. at 5% level	7.95	0.92	4.77	n.s.
at 1% level	11.15	1.28	6.69	n.s.

[a] In pounds of the actual toxicant per acre.

Procedure.—In 1956, field tests were conducted at two locations in Fayette County and at one location in Putnam County, Georgia. Each test involved the application of granulated insecticides to crimson clover in the prebloom stage. The treatments were replicated four times in a randomized block design. The plots were one-fiftieth acre in size and were separated by alleys 10 feet wide.

The insecticides tested were 2% aldrin, 5% dieldrin, 2.5% heptachlor, and 20% toxaphene. All of the insecticides were formulated on 30/60 mesh Attaclay granules and were applied with small rotary dusters. The application dates were March 28 in Putnam County and March 30 at both locations in Fayette County.

The effectiveness of the treatments was evaluated by comparing the numbers of insects caught by sweeping. Ten sweeps with a 12-inch sweep net were taken at random in each plot. Further evaluation was made with seed yields from samples taken by hand-pulling all seed heads from a one-square-yard area selected at random. The seed were threshed in the laboratory with a small electric seed cleaner.

In 1957, experiments were conducted at the following three locations in Georgia: Fayette, Putnam, and Spal-

[1] Paper No. 326; Journal Series, Georgia Experiment Station, Experiment. Accepted for publication January 16, 1958.

Table 2.—Control of weevils on crimson clover with different rates and application dates of two insecticides. Fayette County and Putnam County, Georgia, 1957.

Treatment	Pounds of Toxicant per Acre	Date Applied	Insects[a] per 10 Sweeps		Pounds of Seed per Acre
			Adults	Larvae	
Fayette County					
Heptachlor	2	Mar. 30	0.75	0.50	566
Heptachlor	1	Mar. 30	2.00	0.25	419
Dieldrin	2	Mar. 30	2.00	0.00	333
Dieldrin	1	Mar. 30	3.75	0.25	336
Heptachlor	1	Apr. 12	0.75	1.00	461
Heptachlor	0.5	Apr. 12	1.25	6.00	315
Dieldrin	2	Apr. 12	1.00	4.50	395
Dieldrin	1	Apr. 12	4.75	6.00	227
Untreated			6.50	7.25	163
L.S.D. at 5% level			3.53	2.79	148
at 1% level			4.78	3.79	200
Putnam County					
Heptachlor	2	Apr. 2	0.50	0	606
Heptachlor	1	Apr. 2	1.25	6	516
Dieldrin	2	Apr. 2	3.00	38	483
Dieldrin	1	Apr. 2	12.00	104	476
Heptachlor	1	Apr. 15	2.25	22	529
Heptachlor	0.5	Apr. 15	4.25	70	485
Dieldrin	2	Apr. 15	3.25	61	566
Dieldrin	1	Apr. 15	24.00	159	503
Untreated			47.00	241	343
L.S.D. at 5% level			7.02	42	114
at 1% level			9.51	60	155

[a] *H. meles* and *H. nigrirostris* combined.

ding counties. The treatments consisted of several rates of granulated 5% dieldrin and 2.5% heptachlor applied on two dates. The early application date was set at about March 30 to coincide with the normal practice of removal of the livestock. The later date, April 15, was set to approximate the normal early blooming date of clover and was considered to be the latest time that insecticides could be applied to avoid insecticide hazards to pollinating insects and damage to the crop by equipment used to apply the insecticides.

The treatments were randomized and replicated four times except in Spalding County, where there were only three replicates. A hand-operated fertilizer spreader was used to apply all treatments. Other procedures were the same as those used in the 1956 experiments.

RESULTS AND DISCUSSION.—Results of the experiments conducted in 1956 are presented in table 1. In each of the three tests, the insect infestation was reduced significantly by aldrin, dieldrin, and heptachlor treatments. Toxaphene treatments reduced the infestations somewhat in each test; however, the reductions were not always significant. Seed yields in plots receiving aldrin, dieldrin, and heptachlor in Test 1, Fayette County (table 1) were significantly better than those of the toxaphene-treated and untreated check plots. The yields in Test 2, Fayette County (table 1) showed about the same order of increase in aldrin, dieldrin, and heptachlor treatments; however, only the yields of the heptachlor-treated plots were significantly different from those of the check. In the test in Putnam County (table 1), aldrin, dieldrin, and heptachlor treatments again resulted in increased yields although they were not significantly different from those of the untreated check in this case. A possible explanation

Table 3.—Crimson clover seed yields following insecticide treatments for the control of weevils. Spalding County, Georgia, 1957.

Treatment	Pounds of Toxicant per Acre	Date Applied	Pounds of Seed per Acre
Heptachlor	2	Mar. 28	224
Heptachlor	1	Mar. 28	270
Dieldrin	2	Mar. 28	178
Dieldrin	1	Mar. 28	196
Heptachlor	1	Apr. 12	206
Heptachlor	½	Apr. 12	203
Dieldrin	2	Apr. 12	210
Dieldrin	1	Apr. 12	228
Untreated			153
L.S.D. at 5% level			57
at 1% level			n.s.

for the smaller differences may be that there was considerably more vegetative growth of clover at the time of insecticide application in the latter two locations. There was probably better distribution of the insectcides on the soil surface at the first location where there was little vegetative growth.

Although aldrin gave good control of the insects in the 1956 tests, the resulting yields were consistently lower than were those of the plots receiving dieldrin or heptachlor.

Results of the experiments conducted in 1957 are presented in table 2 and 3. All of the insecticidal treatments reduced the weevil population, with the early applications giving somewhat better control than the later applications. Seed yields were increased following all of the insecticide treatments; however, there were no significant differences in yield due to time of application. Heptachlor gave slightly better yields than dieldrin when compared at the same rate and date of application, although the differences were not significant in most cases.

In 1956, the mean yield of plots which received heptachlor was 889 pounds per acre for the three locations. This represented a 67% increase over the check. In 1957, the highest mean yields for the three locations were 465 pounds per acre on plots receiving 2 pounds of heptachlor per acre and 401 pounds per acre on plots receiving 1 pound of heptachlor per acre. These yields were 112% and 83% greater than the check, respectively.

REFERENCES CITED

Beckham, C. M. 1956. The effectiveness of several insecticides and formulations in the control of seed weevils on crimson clover. Jour. Econ. Ent. **49**(4): 542–4.

Detwiler, John D. 1923. Three little known clover insects. Cornell Univ. Agric. Expt. Sta. Bull. **420.**

Hansen, H. L., and C. K. Dorsey. 1957. Effects of granular dieldrin and heptachlor on adult weevil populations in red clover. Jour. Econ. Ent. **50**(2): 224.

Johnson, W. C., and W. C. Nettles. 1953. Demonstrations in control of clover weevils, 1952 results. Clemson Agric. College Ext. Serv. Misc. Publ. (Unnumbered).

Control of Mites in Laboratory Cultures of the Eye Gnat, *Hippelates Collusor* (Townsend)[1]

MIR S. MULLA,[2] *University of California Citrus Experiment Station, Riverside*

ABSTRACT

Two mite species, one belonging to the genus *Anoetus* (Anoetidae) and the other to the family Pyemotidae, appeared in a laboratory colony of the eye gnat, *Hippelates collusor* (Townsend). The hypopi or deutonymphs of the *Anoetus* species attached themselves to the emerging gnats and made the laboratory-reared gnats worthless for breeding, biological, and toxicological studies. Similarly, the adults of the Pyemotid species, by parasitizing the gnats, rendered them unfit for experimental purposes. Several acaricides were tested for the purpose of controlling these mites without doing appreciable damage to the gnats. Tedion, at a dosage of 50 gm. of 0.1% dust per cage of the larval medium, was the only acaricide among those tested that gave the desired results.

Numerous instances of parasitic and nonparasitic mites, belonging to diverse groups of Acarina, have been reported as infesting insect cultures in the laboratory (Cunliffe 1952, Fisk 1951, Seal & Eden 1956, Baker *et al.* 1956). Once introduced into a colony, such mites build up in large numbers, thus decreasing the value of the culture for experimental purposes. Insects in a colony infested with mites are also unfit for the continuation of the culture.

Fisk (1951) obtained satisfactory control of a species of mite in an American cockroach culture by treating the cockroaches and the inside of the rearing containers with a 5% ovex (Ovotran) dust and spraying the outside of the containers with a suspension of 5% ovex (Ovotran) wettable powder. Baker *et al.* (1956) suggested that one application of DMC (Dimite) or ovex (Ovotran) might control the mite *Machrocheles muscadomesticae* (L.) in house-fly cultures. Seal & Eden (1956) conducted limited studies on the occurrence of three species of mites in commercial cultures of the house cricket, *Acheta domestica* (L.). DMC (Dimite), Aramite, ovex (Ovotran), and Karathane were found to manifest no appreciable toxicity when sprayed at various dosages directly on the insects. However, the effectiveness and practical dosages of these acaricides for the control of these mites were not determined.

In the laboratory colonization of the eye gnat, *Hippelates collusor* (Townsend) (Mulla & Barnes 1957), two species of mites, one of the genus *Anoetus*[3] and the other of the family Pyemotidae, caused considerable inconvenience in the rearing procedure.

Adults of the *Anoetus* species feed on organic matter in the wet larval medium of the eye gnat. Eggs are laid in this medium, and the immature stages spend most of their life cycle there. However, the deutonymphs or hypopi move to the surface of the medium and attach themselves with their suckers to the emerging adult eye gnats (fig. 1) for the purpose of obtaining a free ride. It is mainly in this way that these mites spread from an old medium to a fresh one.

The immature stages of the Pyemotidae species develop in the larval medium, and the adults attach themselves to the sutural and membranous areas of the gnats, particularly around the neck region. It is possible that this mite has a parasitic relationship to the insect. Gnats infested with these mites die soon and are unfit for toxicological and biological studies. Because of the high reproductive capacity of the mites, heavily infested gnats cannot be used for breeding purposes.

FIG. 1. Females of the eye gnat, *Hippelates collusor* (Tsnd.), infested with hypopi of a mite species belonging to the genus *Anoetus*. Upper: freshly emerged. Lower: few hours later with the mites attached to the wings and other parts of the body.

Surface treatment of the medium just at the beginning of the emergence of the gnats with 50 ml. of a 1% spray

[1] Paper No. 1024, University of California Citrus Experiment Station, Riverside, California. Studies conducted under a grant-in-aid from the Coachella Valley Mosquito Abatement District, Riverside County, California. Accepted for publication January 23, 1958.

[2] The assistance of Norman A. Lewis in these studies is acknowledged.

[3] Identified by Dr. Irwin M. Newell, Division of Life Sciences, University of [illegible]ifornia, Riverside.

Table 1.—Acaricides tested against mites (*Anoetus* sp.) breeding in laboratory colony of the eye gnat, *Hippelates collusor* (Townsend).

Material	Concentration (Per Cent)	Amount per Cage (Mixed in Medium)	No. of Cages Treated	Level of Gnat Emergence[a]	Average no. of Mites per Gnat[b]
Dimite	1.0 spray	20 ml.	2	200–500	313
	1.0 dust	100 gm.	3	5–100	0
	0.1 dust	50 gm.	1	200–500	231
Aramite+sulfur	0.016 dust 0.150 dust	100 gm.	2	500–1000	581
Sulfur	1.0 dust	100 gm.	1	500–1000	371
	0.1 dust	50 gm.	1	1000+	342
Ovex	1.0 dust	100 gm.	2	200–500	0
	0.1 dust	100 gm.	2	500–1000	267
Hercules AC-528	0.1 dust	100 gm.	2	0	—
Chlorobenzilate	0.1 dust	100 gm.	3	1000+	641
Tedion	0.1 dust	100 gm.	4	1000+	0
		50 gm.	4	1000+	0
		25 gm.	8	1000+	0
Untreated			19	1000+	458

[a] Level of emergence is an indication of the effect of the chemicals on the immature stages of the gnats.

[b] Mites counted on 15 to 26 gnats per cage where infestation was heavy. More than 200 gnats were inspected where there was no apparent infestation.

of Dimite (prepared from 25% emulsifiable concentrate) gave satisfactory though not complete control of the mites (Mulla & Barnes 1957). At times, two or three such applications were necessary to obtain a large number of gnats relatively free of mites. The timing of the treatment was critical, however, and the degree of control obtained did not result in the elimination of the mites from the colony. The present study was undertaken to evaluate the effectiveness of various acaricides mixed in the larval medium.

METHODS AND MATERIALS.—Wettable-powder formulations of the various acaricides used were diluted with bentonite to obtain dusts containing 1% of each acaricide. These dusts were further diluted with bentonite to prepare more dilute (0.1%) dusts. An Aramite-sulfur dust containing 0.016% Aramite and 0.15% sulfur was also prepared.

Either 100 or 50 gm. of either concentration of the dusts was mixed with 6.5 pounds dry weight of the larval medium mix (5 lbs. vermiculite No. 4, 1.2 lbs. alfalfa meal, and 0.3 lb. Brewer's yeast). After mixing, 2 gallons of water were added to the mixture, and the wet medium was transferred to a rearing cage. Adult eye gnats from cages, the surface of which was treated with Dimite spray at the time of the emergence, were introduced into the rearing cage for egg laying. To expose the damp medium for egg laying it was necessary to disturb the surface of the medium at intervals with a brass rod bent and flattened at one end. This procedure also increased the chances of the transfer of the mites from one cage to another. Untreated cages (usually two to three) were prepared concurrently with the treated ones and were stocked with gnats from the same source.

At the time of the emergence of the gnats, 15 to 26 gnats per cage from the heavily infested cages were transferred into alcohol. The attached mite hypopi were teased off and counted. Two hundred gnats per cage were inspected for hypopi under a dissecting scope in cases where the infestation by the mites was very light or nonexistent.

After preliminary testing, two criteria were established to determine the suitability of an acaricide for incorporation in the larval medium: (1) the acaricide in question should have no appreciable toxicity to the gnats when used at the higher dosage, and (2) it should be relatively effective against the mites at the lower dosage. An acaricide that did not meet these requirements was eliminated from further laboratory trials.

RESULTS.—Results of the tests are presented in table 1. Kelthane at a dosage of 100 gm. of 0.1% dust proved to be toxic to the ovipositing gnats. Further tests with 50 gm. of the 0.1% dust showed similar results. Therefore, under the prescribed criteria of evaluation, no data were obtained on the effectiveness of this material against the mites.

Dimite at a dosage of 100 gm. of 1% dust gave good control of the mites but proved to be toxic to the immature stages of the gnats. Lower dosages of this chemical (20 ml. of 1% spray and 50 gm. of 0.1% dust) were ineffective against the mites and were also toxic to the gnats. Ovex at the higher dosage (100 gm. of 1% dust) controlled the mites but reduced gnat emergence, while at the lower dosage (100 gm. of 0.1% dust), it proved ineffective against the mites and showed slight toxicity against the gnats. Hercules AC-528 proved to be toxic to the gnats at 100 gm. of 0.1% dust, and mite infestation could not be determined.

Sulfur and Aramite-sulfur combination did not show promising results. Chlorobenzilate at a dosage of 100 gm. of 0.1% dust did not control the mites and was not toxic to the gnats.

Of all the acaricides tested, Tedion was the only one that showed effectiveness against the mites with no apparent toxicity to the gnats, at various dosages (table 1). This material was used routinely at a dosage of 50 gm. of the 0.1% dust per cage to eliminate the mites from the colony.

Currently, 10 gm. of 0.1% Tedion dust is incorporated periodically in the larval medium as a precaution against the establishment of future mite infestations. Since the initial elimination of both species of mites from the colony, mite-free gnats have been reared for several generations. Use of this acaricidal treatment is especially desirable when field-collected gnats are used for experimental purposes in the laboratory. A very small number of the wild gnats have been observed to carry one to five mites on their bodies. These few mites, however, are sufficient to establish an infestation under laboratory conditions where food, temperature, and moisture are optimum.

REFERENCES CITED

Baker, E. W., *et al.* 1956. A manual of parasitic mites of medical and economic importance. National Pest Control Association, Inc. New York, N. Y. 170 pp.

Cunliffe, F. 1952. Biology of the cockroach parasite, *Pimeliaphilus podapolipophagus* Träg. With a discussion of the genera *Pimeliaphilus* and *Hirstiella*. Proc. Ent. Soc. Wash. **54**: 153–69.

Fisk, F. W. 1951. Use of a specific mite control in roach and mouse cultures. Jour. Econ. Ent. **44**: 1016.

Mulla, M. S., and M. M. Barnes. 1957. On laboratory colonization of the eye gnat, *Hippelates collusor* (Townsend). Jour. Econ. Ent. **50**(6): 813–6.

Seal, W. L., and W. G. Eden. 1956. Mites in cricket cultures and the toxicities of several acaricides and insecticides to the house cricket. Jour. Econ. Ent. **49**(2): 262–3.

Granulated Insecticide Formulations for Wireworm Control[1]

R. A. Didriksen[2] and J. H. Lilly,[3] *Iowa State College, Ames*

Abstract

This was a laboratory investigation in which wireworms (*Melanotus communis* Gyll.) were exposed to granulated formulations of soil insecticides in quart-size Mason jars. The insecticides employed were aldrin and heptachlor (in separate experiments), and the carriers were No. 4 Vermiculite and 30-60 mesh attapulgus clay. Insecticide concentrations were 2, 5, 10 and 20% on both carriers, plus 30 and 40% on Vermiculite, and the insecticide dosages were 1.25, 2.5 and 5 pounds per acre. Final readings were made 21 days after the experiments were started.

The following conclusions were reached as the result of this work. (1) Granulated Vermiculite was a very satisfactory carrier for both soil insecticides under the conditions of these experiments; No. 4 Vermiculite compared favorably with 30-60 mesh RVM-AA Attaclay. (2) When Vermiculite was used as the carrier for either aldrin or heptachlor there was some decrease in insecticidal efficiency as the concentration of toxicant increased, but the differences between concentrations were not significant in either experiment. (3) With the Attaclay carrier the insecticidal formulations showed significant decreases in insecticidal efficiency as the concentration of toxicant increased, and the number of particles correspondingly decreased, in both experiments.

Since the introduction of granulated insecticides by Whitehead (1952) these formulations have found many uses, one of them being for soil insect control. They eliminate the need for water, enable the farmer to make his own insecticide-fertilizer mixtures, and give him safer formulations. Various materials may be used as granulated carriers. The evaluation of expanded mica (No. 4 Vermiculite) as a carrier for soil insecticides was a major objective of this work. A 30-60 mesh attapulgus clay (RVM-AA Attaclay) was used as a basis of comparison.

Materials and Methods.—Laboratory tests were carried out in quart Mason jars using the wireworm, *Melanotus communis* Gyll., as the test insect. Aldrin and heptachlor were the insecticides used. Different concentrations of the toxicants were used on each carrier to evaluate the effect of varying the number of carrier particles per unit of soil while keeping the amount of actual toxicant constant. Number 4 Vermiculite has a very low bulk density (10 to 12 pounds per cubic foot), and contains approximately 27,500,000 particles per pound. The 30-60 mesh RVM-AA Attaclay carrier has a relatively high bulk density (32 to 34 pounds per cubic foot), and has about 12,300,000 particles per pound. These values are for the unformulated carriers. See table 1 for data on approximate numbers of granules per pound of soil required to give the various aldrin treatments used. The corresponding heptachlor data are so similar that they are not reported.

Aldrin and heptachlor were employed at 1.25, 2.5 and 5 pounds per acre, using concentrations of 2, 5, 10 and 20% of the respective toxicants on both carriers. Also both toxicants were used at 30 and 40% concentrations on Vermiculite. The insecticide dosages were calculated on a weight basis, using 2 million pounds as an average weight of an acre of soil to a depth of 6 inches. (See table 1.) The granulated insecticides were mixed with screened Clarion clay loam soil in quart jars containing 1 pound of soil each. A motor-driven rotary mixer was used to insure even mixing of the insecticides with the soil. Because of the large number of entries separate experiments were run with the two insecticides.

Autoclaved corn was placed near the bottom of each jar as food to help prevent cannibalism. In the heptachlor experiment five wireworms were placed on the surface of the soil in each jar and allowed to work their way into the soil; in the aldrin test the five larvae were placed under the soil surface in each jar. Because of the different timing and procedure it is not possible to make a direct comcomparison between the two toxicants. Each test consisted of three jars or replicates containing five insects each for each treatment.

On the 14th day the jar contents were removed and the wireworms in each were classified as follows: (1) alive (normal in color and at least reasonable active), (2) moribund (abnormal in color and movements feeble and irregular), and (3) dead (showing no movement). A few individuals not recovered were added to the latter group because it was presumed that they had either died and decomposed or deteriorated to the point that they were easy victims of cannibalism. In tabulating the data the moribund larvae were grouped with the dead ones because all our work has indicated that such individuals never recover.

After examination on the 14th day the soil, wireworms and fresh seeds were replaced in the jars and the soil moisture contents brought back to the original level. On the 21st day the wireworms again were examined as above

[1] Accepted for publication January 23, 1958. Journal Paper No. J-3326 of the Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa Project No. 1096. This work was supported in part by a grant-in-aid from the Zonolite Company of Evanston, Ill. Grateful acknowledgment is made to Dr. T. W. Horner, formerly of the Iowa State College Statistical Laboratory, for his assistance with the statistical aspects of this paper.

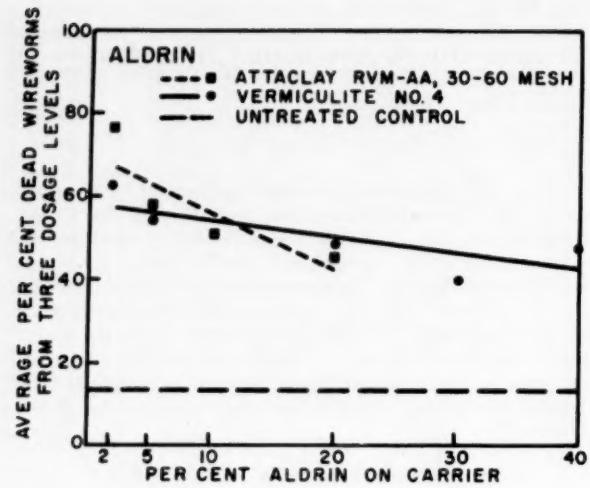
[2] Now Entomologist with the Shell Chemical Corporation.

[3] Now Professor and Head, Department of Entomology and Plant Pathology, University of Massachusetts, Amherst.

Table 1.—Approximate numbers of aldrin-impregnated granules added to 1 pound of soil to give the various dosages used in wireworm tests.[a]

Intended Dosage of Actual Aldrin per Acre	Carrier	Per Cent Concentration of Aldrin on Carrier					
		2	5	10	20	30	40
1.25	Attaclay	394	150	70	30	—	—
1.25	Vermiculite	840	323	151	65	37	22
2.50	Attaclay	787	300	140	61	—	—
2.50	Vermiculite	1681	645	303	131	74	45
5.00	Attaclay	1575	600	280	121	—	—
5.00	Vermiculite	3361	1290	605	261	147	90

[a] These values are based on actual counts made independently by the senior author and Mr. Ray E. Rothfelder of the Zonolite Research Laboratory, Evanston, Ill.


ALDRIN
ATTACLAY RVM-AA, 30-60 MESH
VERMICULITE NO. 4
UNTREATED CONTROL
AVERAGE PER CENT DEAD WIREWORMS FROM THREE DOSAGE LEVELS
PER CENT ALDRIN ON CARRIER

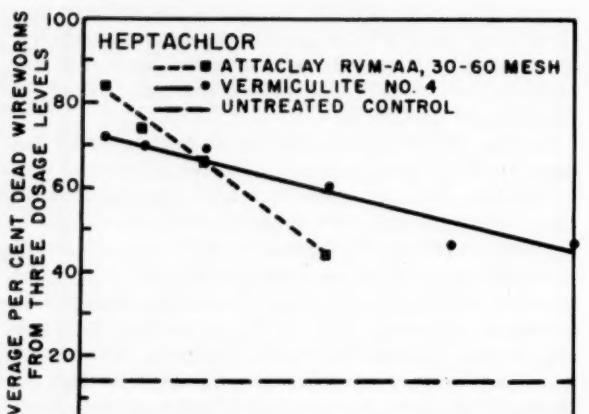

HEPTACHLOR
ATTACLAY RVM-AA, 30-60 MESH
VERMICULITE NO. 4
UNTREATED CONTROL
AVERAGE PER CENT DEAD WIREWORMS FROM THREE DOSAGE LEVELS
PER CENT HEPTACHLOR ON CARRIER

FIG. 1.—Percentages of dead wireworms after 21-day exposures to soil treated with aldrin (top) and heptachlor (bottom). Each point represents the pooled results of three replications of three dosage levels (1.25, 2.5, and 5.0 pounds of actual insecticide per acre).

and the test was terminated. Since the data from the 14th and 21st day examinations showed similar trends, only those of the latter date are included here.

RESULTS.—A separate analysis of variance was calculated on the results of each test. The analyses showed that the differences between the treatments and the controls were significant at the 1% level in both cases. In both experiments the differences between concentrations where Attaclay was used as the carrier were significant (1% level in the heptachlor experiment and the 5% level for aldrin), whereas the differences between concentrations where Vermiculite was used were not significant. In both cases these differences showed linear relationships.

Table 2.—Summary of wireworm mortalities after 21 days of exposure to soil treated with two granulated insecticides at three dosage rates. Iowa—1955.[a]

Carrier and Per Cent Insecticide Concentration		Total Wireworm Mortality[b]					
		Aldrin			Heptachlor		
		1.25	2.5	5.0[c]	1.25	2.5	5.0[c]
Attaclay	2	11	8	15	11	12	15
Vermiculite	2	8	9	11	7	10	15
Attaclay	5	5	9	12	9	11	13
Vermiculite	5	6	7	11	9	9	13
Attaclay	10	5	7	11	7	9	14
Vermiculite	10	4	11	10	9	9	13
Attaclay	20	5	8	8	3	6	11
Vermiculite	20	2	8	12	3	11	13
Vermiculite	30	2	5	11	4	6	11
Vermiculite	40	5	7	10	8	5	9
Untreated control		2	2	2	3	1	2

[a] The results obtained with the two insecticides were from two separate experiments and hence are not directly comparable.
[b] Total of three replicates of five wireworms each (15 individuals) for each carrier and each insecticide concentration.
[c] These figures represent the pounds of actual insecticide per acre, calculated on the assumption that the top 6 inches of an acre of soil weighs 2,000,000 pounds.

The relative differences between concentrations were the same for the three dosage levels in both experiments, *i.e.*, the interaction between concentration and dosage level was not significant with either insecticide. Therefore the results obtained from the three dosage levels were pooled to give the means shown in figure 1. The data on which the points shown in this figure are based are presented in table 2.

Figure 1 shows that, when the amount of toxicant remained constant, decreasing the concentration (*i.e.*, increasing the number of granules per unit of soil) resulted, in general, in increased wireworm mortality. Since all factors except the insecticide concentration or the number of granules per unit area remained the same, it follows that the differences in wireworm mortality were due primarily to the numbers of granules employed. Apparently in the higher toxicant concentrations on Attaclay the numbers of granules per unit of soil were approaching the lower limit necessary for good control, whereas with the Vermiculite formulations there were sufficient granules to give control.

REFERENCES CITED

Whitehead, F. E. 1952. Large scale experiments in rice field mosquito control. Arkansas Agric. Expt. Sta. Report Service **32**: 1–15.

Food Habits of the Khapra Beetle Larva[1]

ZENAS B. NOON, JR.[2]

ABSTRACT

Khapra beetle larvae, *Trogoderma granarium* Everts, feed on foods other than on grains and related products. Experiments show that these larvae, like those of many other dermestids, feed on substances of animal origin. Elimination-type preference tests demonstrated that larvae prefer them to many vegetable foods. This indicates that complete eradication of the beetle may not be possible unless such food reservoirs are taken into consideration. The feeding tests also showed, contrary to previous statements, that first instar larvae can attack sound grain and survive.

The khapra beetle, *Trogoderma granarium* Everts, has caused extensive damage to stored grain since its recent discovery in the southwestern United States. To bring the pest under control, a knowledge of its food preferences and feeding habits is essential. Previous investigations have been limited mainly to studies involving late instar larvae. Reports on the ability of first instar larvae to attack, and survive in, sound grain are conflicting. Feeding capabilities of the larvae and the foods on which they might survive are also not clear.

Most authors state that larvae can penetrate the grain only after the third molt (Voelkel 1924, Barnes & Grove 1916, Lepesme 1938, Hinton 1945), but Hadaway (1956) reports that "young larvae" can apparently attack whole grain by biting through the bran. Hopkins (1955) states, that in general, larvae prefer cracked grain and grain dust to clean, sound grain. Although it is believed that khapra beetle larvae prefer cereal products to substances of animal origin, some authors report that they will develop on dead mice, dried blood, dried insects and similar animal material (Hinton 1945, Harper 1955). It is widely known that the larvae will develop on grains such as wheat, rye, oats, barley, rice and processed cereal products. Hopkins (1955) states that larvae develop faster on processed and milled products than on unprocessed materials. Results of actual tests concerning larval food preferences have not been published. The stated preferences for cereal products over animal material are apparently opinions of the authors involved. A study of other possible food materials occurring in regions of infestation and which might serve as natural reservoirs, seemed desirable. A knowledge of such reservoirs would then be of value in control operations against the pest.

MATERIALS AND METHODS.—A variety of animal and vegetable matter was tested. Animal matter consisted of dead insects and other arthropods (table 1), dead vertebrates and other animal matter (table 1). The vegetable matter consisted of grain, seeds and milled products (table 2), and miscellaneous plant products (table 2).

The khapra beetles used in the experiments were obtained from infested grain bins in Phoenix, Arizona. The adults were identified as *Trogoderma granarium* Everts by Dr. R. S. Beal, Jr. Adult beetles, taken from cultures, were placed in small stender dishes. Eggs laid in the dishes were checked daily and any first instar larvae found were transferred to 10-centimeter petri dishes containing one of the test foods. The food dishes were inspected frequently. Dead larvae or adults were removed so they would not be eaten by the remaining larvae. The tests were run from July to December at seasonal temperatures in an uninsulated shed. A laboratory oven, set at 35° C., was employed thereafter to hasten larval development and finish the experiments.

RESULTS.—Results of the feeding tests are summarized in tables 1 and 2. The tables show the number of larvae started on the food, number of adults emerging, days to maturity, number of larvae dying and number remaining at the close of the experiments. Although khapra beetle larvae prefer vegetable matter, the experiments show that like other members of the dermestid family, they will feed on matter of animal origin. The larvae developed rapidly on dead insect remains. Development on honeybee remains was slow and most larvae died; however, some late instar larvae were still alive at the close of the experiment. The hairiness of the honeybee may have been a factor which hindered feeding of the earlier instars.

Development on dead vertebrates was more difficult for the larvae. At the close of the experiments they had com-

[1] Arizona Agricultural Experiment Station Technical Paper No. 445. Accepted for publication January 24, 1958.

This work was carried out under the terms of Contract 12-25-010-96 between the Board of Regents of the University and State Colleges of Arizona and the Stored-Product Insects Section, Agricultural Marketing Service, United States Department of Agriculture. The Agricultural Marketing Service neither restricts nor is responsible for the opinions or conclusions of the author.

[2] Present address: Department of Entomology, University of Illinois, Urbana, Illinois.

Table 1.—Summary of experiments in which first instar khapra beetle larvae were placed on animal matter.[a]

Food	Larvae Started	Adults	Days to Maturity	Larvae Died	Larvae Remained
Dead insects and other arthropods					
Wasp nest with eggs, larvae and pupae, *Polistes exclamans arizonensis* Snelling	56	27	33–204	26	3
Apis mellifera L.	32	—	—	29	3
Polistes exclamans arizonensis Snelling	12	2	195, 196	8	2
Moths	50	6	128–158	42	2
Khapra beetle adults	50	—	—	50	—
Schistocerca sp., *Boopedon* sp.	24	6	34–201	17	1
Miscellaneous insects[b]	26	3	42–157	15	8
Scorpions	21	4	157–231	17	—
Dead vertebrates and other animal matter					
Dried meat of ground squirrel, *Citellus* sp.	61	—	—	61	—
Lizard, *Sceloporus* sp.	53	4	151–182	45	4
Dried meat of deer, *Odocoileus hemionus* Merriam	50	—	—	21	29
Dried meat of quail, *Lophortyx gambeli* Gambel	56	2	188, 207	48	6
Cattle hair	50	—	—	50	—
Raw wool	61	—	—	61	—
Parmesan cheese	60	—	—	60	—
Honeycomb	59	—	—	59	—
Beeswax	61	—	—	61	—
Powdered milk	64	—	—	39	25
Dung (cattle)	57	—	—	57	—
Dung (cattle)[c]	50	—	—	50	—

[a] The experiments were run at seasonal temperatures in an uninsulated shed from July to December. A laboratory oven, set at 35° C., was employed thereafter to hasten larval development and finish the experiments.

[b] Robber flies, June beetles, and ground beetles.

[c] Late-instar larvae used in this test.

Table 2.—Summary of experiments in which first instar khapra beetle larvae were placed on vegetable matter.[a]

Food	Larvae Started	Adults	Days to Maturity	Larvae	
				Died	Remained
Grain, seeds, and milled products					
Sound milo grain	51	1	170	50	—
Cracked milo grain	55	18	151–187	37	—
Sound barley grain	57	—	—	57	—
Cracked barley grain	50	—	—	50	—
Puncture vine seeds, *Tribulus terrestris* L.	53	—	—	53	—
Golden crownbeard seeds, *Verbesina encelioides* (Cav.) Benth. and Hook.	10	—	—	10	—
Mesquite beans, *Prosopis juliflora* (Swartz) var. *velutina* (Woot.)	55	—	—	55	—
Flour (white)	56	20	138–219	31	5
Oatmeal	50	20	152–288	—	30
Corn meal (yellow)	52	27	125–276	6	19
Miscellaneous plant products					
Leafcutter bee provisions, *Megachile* sp.	49	23	132–208	26	—
Honeycomb with pollen	12	7	38–163	—	5
Pollen balls of honeybee	52	30	37–207	22	—
Propolis of honeybee	53	—	—	53	—

[a] The experiments were run at seasonal temperatures in an uninsulated shed from July to December. A laboratory oven set at 35° C., was employed thereafter to hasten larval development and finish the experiments.

pleted their growth only on quail and lizard remains. Tests still in progress show that the larvae are developing slowly on deer meat, but have reached a stage of growth where they should mature. None of the larvae survived on ground squirrel remains, the meat of which was very oily. They were not able to move about and feed on this food and eventually became trapped in the oil.

The larvae were unable to survive on cattle hair, raw wool, honeycomb, beeswax, dung or Parmesan cheese. Larvae were seen feeding on cattle dung, cattle hair, raw wool, honeycomb and beeswax, but perhaps because of a lack of certain essential nutrients they were unable to survive for longer than 9 days. Although some development was possible on cheese, eventually all larvae died. The oily nature of the cheese created feeding difficulties similar to those encountered with squirrel meat. Dried-milk experiments have been in progress only a short time, but it is evident from the larval growth that their development will be completed.

These food tests confirm previous beliefs that khapra beetle larvae are, in most cases, able to complete their development on grain and milled products. Of 51 first instar larvae started on sound milo kernels, one completed its development. This test on uncracked milo shows that larvae are capable of penetrating and feeding on this particular grain, at least, prior to the fourth instar. This evidence is contrary to previous information on the ability of young larvae to penetrate sound grain (Voelkel 1924, Barnes & Grove 1916, Lepesme 1938, Hinton 1945). Progress beyond the first instar, however, was impossible with either sound or cracked barley.

Larvae placed on puncture vine seeds, golden crownbeard seeds and mesquite bean pods were unable to complete their life cycles. With puncture vine seeds and mesquite beans, larvae had difficulty in penetrating the hard seed coats, although a few larvae reached the third instar on pods of the latter. The pubescence of the golden crownbeard seeds apparently prevented small larvae from attacking the seed coat itself.

Table 3.—Summary of data from elimination-type food preference tests with the khapra beetle.[a]

Food Material	Days Feeding	Food Material	Days Feeding	Preference
Pollen	1	Cracked milo	39	Cracked milo
Pollen	35	Cracked barley	4	Pollen
Dead insect	40	Cracked milo	0	Dead insect
Dead insect	36	Cracked barley	1	Dead insect
Cracked barley	3	Cracked milo	31	Cracked milo
Pollen	0	Dead insect	39	Dead insect
Pollen	0	Deer meat	40	Deer meat
Pollen	2	Oatmeal	36	Oatmeal
Cracked milo	1	Deer meat	39	Deer meat
Cracked milo	2	Oatmeal	38	Oatmeal
Deer meat	39	Cracked barley	0	Deer meat
Cracked barley	8	Oatmeal	29	Oatmeal
Dead insect	3	Deer meat	35	Deer meat
Dead insect	21	Oatmeal	16	Dead insect
Deer meat	39	Oatmeal	1	Deer meat
Dead insect[b]	38	Oatmeal	2	Dead insect

[a] The experiments were run at seasonal temperatures in an uninsulated shed.
[b] Re-test between dead insect and oatmeal.
Preferences are as follows: 1, deer meat; 2, dead insect; 3, oatmeal; 4, cracked milo; 5, pollen; 6, cracked barley.

Of all the vegetable products tested pollen seemed to furnish the most ideal diet. Almost half of the larvae placed on pollen completed their life cycles. This was well over the proportion that completed life cycles on any of the other vegetable products tested.

Preference Tests.—The feeding experiments demonstrated that khapra beetle larvae feed and develop almost equally well on either vegetable or animal matter. The time required for this insect to complete its life cycle was about the same on both types of food. Hence, a food preference could not be determined from those experiments. Elimination-type preference tests were set up to determine actual preferences between different types of food. Foods used in this test represented the different categories of vegetable and animal matter used in the feeding experiments: cracked milo, cracked barley, oatmeal and pollen; dead insects (carabid beetle), and dried deer meat. Each food was paired once with each of the other foods so that all possible combinations (15) were obtained. Each food combination was placed in a separate petri dish with 10 nearly mature larvae. The foods were placed in separate piles on opposite sides of the dish. The dishes were checked daily for 40 days and records were kept of the number of larvae feeding (or not feeding) on each food. A summary of food preferences is given in table 3.

A non-parametric analysis was used for evaluating the data. The food on which the most larvae were feeding was given the preference for the day. The results of each daily test were then tabulated to determine which of the foods was preferred. In all tests except the one between oatmeal and dead insect, the decision was considered significant. The test on the oatmeal and dead insect phase was repeated.

The experiments established a definite sequence of food preferences as follows: deer meat, dead insects, oatmeal, cracked milo, pollen and cracked barley (table 3). These simple tests, although not conclusive, do suggest that khapra beetle larvae prefer foods of animal origin. Processed cereal products appear to be favored slightly over unprocessed products.

DISCUSSION.—Previous feeding experiments have been conducted on vegetable matter, but the larvae used in them were all of late instars (Hopkins 1955). To insure

accuracy, most of the present experiments were started by placing only first instar larvae on the foods tested. The experiments described here were set up to determine the range of food types on which the khapra beetle larvae might feed and develop. A further question to be answered by these feeding experiments was whether or not khapra beetle larvae younger than the fourth instar could penetrate sound grain.

It was found that khapra beetle larvae, like many of their close relatives, feed and survive on animal matter. The larvae developed rapidly on dead insect material. Their development was somewhat slower on other animal material, such as dead birds and animals. Experiments being conducted with powdered milk indicate that the larvae are developing satisfactorily and may be expected to mature. Larvae placed on cattle hair and raw wool were unable to survive. Some dermestids prefer this type of food, but it is not adequate for the khapra beetle. Similarly, larvae were unable to develop on cattle dung, regardless of the stage of growth they had previously attained.

The food experiments demonstrated that larvae will feed and develop on several types of vegetable matter. Certain types of seeds, however, such as mesquite beans, golden crownbeard seeds and even sound and cracked barley were impenetrable to first-instar larvae.

Barley was the least preferred of all the foods, according to elimination-type food preference tests (table 3). A survey of khapra beetle infestations in Arizona seemed to indicate that barley was its preferred food. About one-third of all Arizona infestations occurred in barley (Nutting 1956). From this evidence it would seem that larvae should be able to develop readily on barley. Failure of first-instar larvae to survive in small cultures, even on cracked barley, suggests that infestations must be started by late instar larvae. Population increases are likely made possible through growth of first instar larvae on grain dust and debris. Nutting (*op. cit.*) suggested that the apparent high infestation rate in barley might result from the grain being widely placed in vulnerable bulk storages and commonly held for periods of longer than 1 year.

The experiment with sound milo showed that larvae younger than the fourth instar are capable of penetrating and surviving on sound grain. It is obvious that even if this particular grain is clean and sound it is still vulnerable to khapra beetle attack.

Hopkins (1955) states that the larvae develop faster on processed and milled products than on unprocessed material. Contrary to his findings, the feeding experiments here show that there is no great difference in the development time required by larvae fed on processed and unprocessed materials. Since Hopkins, in his tests, used larvae which were slightly less than half grown, some larvae may have been older than others and therefore matured faster. The present experiments, which were all started with first instar larvae, provide a better evaluation of the suitability of the foods tested. Hopkins' experiments may indicate that late instar larvae can develop faster on some products, but there was no such evidence in the present experiments.

The elimination-type food preference tests show that of all the materials used the khapra beetle larvae prefer animal matter to vegetable matter. This is contrary to previous reports that larvae prefer cereal products to those of animal origin (Hinton 1945, Harper 1955). It is possible that the khapra beetle, long known as a stored-grain pest, has gone unnoticed on many animal products. With the knowledge from these food and elimination-type preference experiments it is indicated that a program directed toward the eradication of the khapra beetle may not be accomplished by eliminating the pest from stored products alone.

It is clear that the larvae can and will develop on a wide variety of materials commonly found in the vicinity of storage areas. Such foods as pollen and dead insects, for example, could provide convenient natural reservoirs from which new infestations might be started.

REFERENCES CITED

Barnes, J. H., and A. J. Grove. 1916. The insects attacking stored wheat in the Punjab, and the methods of combating them. India Dept. Agric. Mem. **4**(6): 165–280.

Hadaway, A. B. 1956. The biology of the dermestid beetles *Trogoderma granarium* Everts and *Trogoderma versicolor* (Creutz). Bull. Ent. Res. **46**: 781–96.

Harper, R. W. 1955. Khapra beetle and Mexican fruit fly. California Dept. Agric. Bull. **44**(1): 17–28.

Hinton, H. E. 1945. A monograph of the beetles associated with stored products. British Museum **1**: 392–5.

Hopkins, L. 1955. Food preferences of the khapra beetle. Jour. Econ. Ent. **48**(3): 332–3.

Howe, R. W., and H. D. Burges. 1956. *Trogoderma afrum* Priesner, a synonym of *T. granarium* Everts and a comparisom with *T. versicolor* (Creutz.). Bull. Ent. Rex. **46**(4): 773–80.

Lepesme, P. 1938. Sur le régime et l'importance économique de quelques *Trogoderma* (Coleoptera Dermestidae). Rev. Franç. Ent. **5** (fasc. 2): 104–106. *Abstracted in:* Rev. Appl. Ent., Ser. A, **27**: 196.

Nutting, W. L. 1956. Personal communication.

Voelkel, H. 1924. Zur Biologie und Bekämpfung des Khaprakäfers, *Trogoderma granarium* Everts. Arb. Biol. Reichsanst. Land- u. Fortsw. **13**(2): 129–71. *Abstracted in:* Rev. Appl. Ent., Ser. A, **12**: 482.

Insecticide Protection Against *Pagiocerus fiorii* in Ecuador[1]

HAROLD R. YUST[2] and MIGUEL A. CEVALLOS G.[3,4]

ABSTRACT

Methoxychlor, lindane, malathion and pyrenone dusts were tested against *Pagiocerus fiorii* (Egger) in stored soft corn. In corn that was less than 1% attacked, methoxychlor at 50 p.p.m. gave protection for over 9 months and in field trials prevented population buildup for 3 months in corn that was less than 10% attacked at the time of treatment. Lindane at 10 p.p.m. and pyrenone at 1 pound per 10 bushels gave protection for 6 to 7 months when the corn was less than 1% infested.

Soft corn grown in the Andean mountains of Ecuador is often destroyed in storage by a Scolytid beetle, *Pagiocerus fiorii* (Egger). The corn is infested in the field before the grain is sufficiently dry to harvest. The authors have seen corn in the warmer valleys that was more than 50% attacked 3 to 4 months after storage. Undoubtedly high populations of the beetle were within flying distance and there were other favorable factors, as described by Yust (1957), to cause such rapid infestation but it clearly shows that farmers have reasons to fear this weevil.

During the course of several years a number of tests were made with insecticides of relatively low toxicity to warm blooded animals to determine the protection afforded stored corn against this pest and to promote the use of the less toxic insecticides at minimum dosages. In Ecuador there are no legal tolerances or restrictions on the use of insecticides on stored grains. This paper reports the results of some tests made in the Quito Laboratory and the results of a few practical treatments made in the field.

METHODS AND PROCEDURES.—The corn used in the tests in the laboratory was obtained 1 to 2 months after harvest in Cumbayá where the beetles are a serious pest. Both corn on the cob and shelled corn were included in the study and were from the same field. The shelled corn was removed from the cob immediately before treatment. Since the corn was stored on the cob it was reasonably dry when treated but the moisture content was not determined. The corn was good quality, was thoroughly mixed before treatment to assure uniform lots in regard to infestation and was given no special exposure to weevils before treatment. The corn was typical of the area.

One sack of 100 pounds of shelled corn and one of 60 pounds of corn on the cob were treated in each instance with one or more dosages of 4 insecticides. One sack each of the same weights was used for untreated checks. The insecticides were applied directly on the corn as dusts and were thoroughly mixed with the grain by hand. All treated and untreated lots were placed at random in the same room where they were held throughout the study. Naturally, the treated corn was exposed to a very high population of weevils as the untreated sacks became heavily infested.

The insect population was determined by the number of attacked kernels in 10 samples of 50 kernels each drawn at random from the sacks. The kernels were observed for evidences of feeding or the presence of larvae and adults. All kernels with feeding lesions or containing insects were recorded as attacked and the average, in percentage of grain attacked, was used as the index of infestation. The first count was made at the time of treatment and others followed at about monthly intervals. The same general system was used for both unshelled corn and shelled corn but the corn on the cob was not removed from the ear to make the inspections.

In field trials large quantities of grain were usually treated with but one insecticide but in two cases the sack method was used as reported for the laboratory tests. The same procedure of making counts was used in both the laboratory and the field but in the latter, counts were not made at regular intervals. Frequently, the untreated lots were treated or sold by the farmers during the intervals between inspections because of obvious infestations, so many records are not complete.

LABORATORY TESTS.—Methoxychlor, lindane, malathion and pyrenone[5] were applied on various lots of corn as indicated in table 1. The dosage of insecticide is given in pounds of insecticide mixture per 1000 pounds of grain. It was assumed that the cob was one sixth of the weight of the unshelled corn in which case the dosage of insecticide was for 1200 pounds of corn on the cob. The dosages are equivalent to 50 p.p.m. of methoxychlor, 5 and 10 p.p.m. of lindane and 8 p.p.m. of malathion. The population during the first six counts was practically zero in all treated

[1] Accepted for publication January 27, 1958.

[2] Entomologist for International Cooperation Administration stationed in Ecuador; now Deputy Director, Division of Agriculture, Lima, Peru.

[3] Entomologist for Servicio Cooperativo Interamericano de Agricultura in Ecuador; now entomologist for Standard Fruit Shipping Company.

[4] The authors wish to thank Sr. Angel Arnulfo Ibarra E. for his assistance in obtaining part of the data contained in this paper.

[5] The active ingredients of the pyrenone that was used are reported to be 0.8% of piperonyl butoxide and 0.05% of pyrethrins and the carrier was reported to be fabrous talc.

Table 1.—Percentage of corn infested with *Pagiocerus fiorii* in the laboratory test at indicated days after treatment.

INSECTICIDE	DOSAGE[a]	PER CENT ATTACKED KERNELS (DAYS AFTER TREATMENT)			
		190	220	251	282
Shelled Corn					
Methoxychlor 2.5%	2	1.0	1.6	1.6	1.0
Lindane 0.25%	2	1.4	2.0	4.0	55.8
Lindane 0.5%	2	1.2	2.0	2.8	37.0
Malathion 0.5%	1.6	1.2	2.0	2.6	1.6
Pyrenone	1.8	1.0	2.2	8.8	62.6
Untreated	—	97.6	99.6	100.0	—
Ear Corn					
Methoxychlor 2.5%	2	1.2	1.8	0.4	1.6
Lindane 0.25%	2	1.4	21.4	42.6	85.0
Lindane 0.5%	2	2.2	25.8	49.8	86.8
Malathion 0.5%	1.6	1.2	1.0	1.0	4.8
Pyrenone	1.8	1.0	8.2	13.8	79.4
Untreated	—	80.2	99.4	100.0	—

[a] Pounds of insecticide mixture per 1000 pounds of corn.

lots so no data are included until about 6 months after treatment. At the time of treatment 0.7% of the shelled corn and 0.3% of the ear corn was infested.

The data clearly show that methoxychlor at 50 p.p.m. protected the corn for more than 9 months in the presence of a heavy population of weevils. Malathion at 8 p.p.m. was about as effective as methoxychlor and both were superior to lindane and pyrenone. The latter two insecticides gave control for 6 or more months which is about the average storage period. The conditions of the laboratory were probably similar to storage bins in the warmer valleys because 78% of the untreated shelled corn was infested 129 days after the test was started and 45% of the unshelled corn was infested during the same interval.

FIELD TRIALS.—Soft yellow corn was treated during November in Sangolqui at the rate of 1 pound of pyrenone per 560 pounds. The corn was less than 1% attacked when treated. Six months later the treated corn had less than 1% of attacked kernels and the untreated had 40.6% of attacked kernels. The same type of shelled corn was treated in San Antonio with pyrenone at the same dosage. Before treatment the corn was 0.8% infested. Two months later the treated corn remained the same and the untreated was 33.2% infested. Many other cases can be cited to show that pyrenone protects stored corn when less than 1% of the corn is infested at the time of treatment.

Tests with 1 pound of 1% lindane per 1000 pounds of corn gave good control at San Antonio when the corn was 0.8% infested before treatment. In San José de Minas the same dosage of lindane was used when 6.8% of the corn was infested. About 4 months later the untreated corn was 90% infested and the treated averaged 64.5% infested. Similar results were obtained with pyrenone at 1 pound per 560 pounds of corn.

Field treatments were made on many occasions in different locations using 2.5 and 5% methoxychlor where farmers reported very serious damage from *P. fiorii*. In tests with ear corn the lower concentration was used at 1 pound per 550 pounds and the higher concentration was used at 1 pound per 1100 pounds. All treatments protected the corn for a period of 5 to 8 months after which time the farmers sold the grain. Both concentrations of the insecticide seemed to give equally good protection.

On various occasions corn that was up to about 10% infested was treated to determine whether the population would be controlled by treatment. One such treatment was made in Perucho with 2.5% methoxychlor at the rate of 1 pound per 500 pounds of corn on the cob. Two thousand pounds were treated and 1000 pounds were left in the same room without treatment. The corn was 7.8% infested before treatment. Three months later the infestation in the treated corn was unchanged and that in the untreated increased to 49.4%. In general the treatments seem to indicate that methoxychlor controls the beetle for a time when the infestation does not exceed 10% before treatment. This is especially important because lindane and pyrenone failed to protect corn when the infestation exceeded 1%.

REFERENCES CITED

Wille, J. E. 1943. Entomologia Agrícola del Perú. Editado por la Estación Experimental Agrícola de la Molina, Ministerio de Agricultura, Lima, Perú.

Yust, H. R., and M. A. Cevallos. 1954–55. Lista preliminar de plagas de la Agricultura del Ecuador. Rev. Ecuat. Ent. Par. 2(3–4); 425–42.

Yust, H. R. 1957. Biology and habits of *Pagiocerus fiorii* in Ecuador. Jour. Econ. Ent. 50(1): 92–96.

Insecticides to Prevent the Emergence of *Scolytus multistriatus*[1]

CHARLES C. DOANE, *The Connecticut Agricultural Experiment Station, New Haven*

ABSTRACT

Emulsions of four insecticides were tested for their effectiveness in preventing emergence of the smaller European elm bark beetle, *Scolytus multistriatus* (Marsh.). Emulsions of 12% DDT, 6% dieldrin and 6% lindane were applied to bark brought into the laboratory. The bark surface was only slightly moistened during spraying. Lindane was most effective, followed by dieldrin and DDT in reducing emergence by 97%, 51% and 22%, respectively. Daily readings of the treatments with lindane, dieldrin, and DDT showed that 1%, 5% and 41%, respectively, of the beetles were alive 24 hours after emerging. Beetles were in the larval stage at time of treatment and none of the insecticides prevented them from reaching the adult stage.

Two replicated field tests were run using 0.25% to 2% sprays of DDT, dieldrin, lindane and malathion in amounts that wetted the bark of the infested elm logs. The beetles were in the larval or pupal stage when treated and again the sprays did not kill the insects until they reached the adult form. All sprays penetrated better when the bark was thoroughly wetted whereas only lindane penetrated well in the laboratory test. Lindane and dieldrin sprays at either 0.5% or 1% were very effective in reducing emergence. DDT sprays were less effective while 0.5% or 1% malathion sprays were least efficient.

The smaller European elm bark beetle, *Scolytus multistriatus* (Marsh.), is one of the principal vectors of the Dutch elm disease pathogen. The most important carriers are those beetles that pass the larval stage in the bark during the winter and emerge as adults in the spring. This is based on the finding that elms are most susceptible

[1] Accepted for publication January 27, 1958.

Table 1.—Per cent mortality of smaller European elm bark beetles emerging from bark treated with insecticide emulsions in the laboratory.

Insecticide	Beetles Killed in Bark			Emerged Beetles[b]	
	Surface[a]	In Pupal Cell	Total	Alive	Dead
DDT 12%	17	5	22	41	37
Dieldrin 6%	42	9	51	5	44
Lindane 6%	29	68	97	1	2
Untreated	5	12	17	76	7

[a] Beetles that left pupal cell and attempted emergence.
[b] Emerged beetles were removed and counted each 24 hours for 34 days following first emergence.

during the spring and early summer and then become relatively difficult to infect after the first of August (Parker *et al.* 1941). The common practice is to remove such breeding material during the winter months. However, tree removal programs are often delayed with the result that many elms with dangerous infestations of elm bark beetles are not destroyed before emergence begins. If insecticides were used to prevent emergence of the beetles the trees would not serve as sources of infection and could be removed later. Working in this direction Whitten (1942) obtained high kills of elm bark beetles with several chemicals including orthodichlorobenzene and naphthalene mixed with fuel oil. Whitten (1945) also found that DDT in kerosene killed all of the larvae in infested bark while DDT in emulsion form only partly prevented emergence. Those beetles that did emerge were unable to construct new brood galleries in untreated logs placed in the same cage. Becker (1950) has indicated that emulsions of chlordane and BHC also show promise. The present paper reports on the toxicity of four chlorinated organic insecticides applied as emulsions to prevent the emergence of smaller European elm bark beetles from the infested bark.

Laboratory Study.—Elm bark known to be infested with larvae of the smaller European elm bark beetle was cut into sections and treated with emulsions of 12% DDT, 6% dieldrin, or 6% lindane. The sprayer used in the test was a modification of a precision laboratory sprayer designed by Hoskins (Turner & Woodruff 1946). It was set to deliver 0.026 ml. of spray per 100 square centimeters of bark surface during the 5-second spray period. The bark became slightly moistened but not wet. The sections were allowed to dry for an hour and were then placed in half-gallon containers. Three containers, each with two pieces of bark, were used for each treatment. These containers were connected in tandem with rubber hose so that moist air could be circulated through each container to prevent accumulation of insecticide vapors.

Beetle emergence began about 20 days after treatment. For the next 34 days all beetles were removed from the containers each 24 hours and classified as dead, moribund or alive. The positive phototropic response of the beetles was used to capture the adults in glass vials attached to each container. This tended to reduce the time that emerged beetles spent on treated bark. When emergence was complete each piece of bark was dissected and the stage and position of insects unable to emerge was recorded. The adults killed in the pupal cell were recorded separately from those that left the cell and attempted to emerge. This was to give some indication of the penetrative ability of the insecticides. No data on the mortality of immature forms are included. The small number that died did so independent of treatment.

Table 2.—Per cent mortality of smaller European elm bark beetles emerging from the bark of logs treated with emulsions of various insecticides.

Insecticide and Per Cent Concentration		March 22 Test			May 6 Test		
		Surface[a]	In Pupal Cells[b]	Total	Surface	In Pupal Cell	Total
DDT	0.5	—	—	—	12	27	39
DDT	1	25	29	54	20	51	71
DDT	2	22	42	64	7	39	46
Dieldrin	0.25	—	—	—	17	41	58
Dieldrin	0.50	19	66	85	23	45	68
Dieldrin	1	13	70	83	26	41	69
Lindane	0.25	—	—	—	14	39	53
Lindane	0.5	12	73	85	17	61	78
Lindane	1	20	74	94	2	70	72
Malathion	0.5	3	19	22	—	—	—
Malathion	1	8	40	48	—	—	—
Untreated	—	0	2	2	0	2	2

[a] Beetles that left pupal cell and attempted emergence.
[b] Beetles killed in the pupal cell.

Results and Discussion.— None of the insecticides prevented the immature insects from becoming adults. However, the response of the adults to the three chemicals tested was markedly different (table 1). Lindane was the most effective in preventing emergence and by penetration was able to kill the greatest number of beetles before they left the pupal cells. Only 3% emerged at all while the remainder of the beetles were killed as they attempted emergence. Dieldrin apparently did not affect the beetles in the pupal cells but was very efficient in killing the beetles after they left the pupal cells to open holes for exit. Many dead adults were found partially exposed in the dieldrin treatments. While 49% emerged from the dieldrin-treated bark, only 5% of all beetles were alive 24 hours after emergence. DDT was not as effective as dieldrin or lindane since 41% were alive 24 hours after emergence. In the field such beetles theoretically could fly to healthy trees and feed; there would be much less chance of this with beetles that die within the first 24 hours.

Field Study.— Two different sets of experimental materials were sprayed. On March 22, 1957, insecticides were applied to the bark of 14-inch elm blocks 4 to 8 inches in diameter. Five blocks were treated with each insecticide formulation. All the beetles in the bark of these blocks were in the larval stage. On May 6, 1957 sprays were applied to the bark of 4-foot lengths of elm logs that were 3 to 7 inches in diameter. Four logs were sprayed with each treatment.

Application methods for both sets were approximately the same. The insecticide emulsions were applied with a 3-gallon pressure-type sprayer. The bark was dry on both

sets of logs before treatment so the sprays soaked in soon after treatment. The spray was applied until the point of run-off began and the logs were then rolled and sprayed until complete coverage was attained. No mortality counts of emerging beetles were made. After beetle emergence was complete samples of bark were removed from each replicate. The number of exit holes were counted as healthy beetles and then the bark samples were dissected in order to record the stage and position of the insects unable to emerge.

Results and Discussion.—Results of both field tests indicated that the insecticides did not prevent the immature stages from becoming adults (table 2). Sprays containing 0.5% or 1% lindane gave the highest kill followed closely by 0.5% and 1% dieldrin. Sprays containing 1% or 2% DDT were less efficient while the malathion sprays were least efficient in preventing emergence. A comparison of the numbers killed in the pupal cells with the numbers of beetles killed during emergence indicated that lindane, dieldrin and DDT all penetrated well in contrast to the results in the laboratory test where only lindane did so. The high gallonage of spray used in the field tests apparently carried the insecticides deeper into the bark.

The field tests did not account for the fate of those beetles able to emerge. From the results of the laboratory test, however, it is reasonable to expect that many of the beetles emerging from treated logs or trees in the field would be unable to fly to healthy twigs.

REFERENCES CITED

Becker, W. B. 1950. Sprays to kill scolytids breeding in elm logs. Massachusetts Agric. Expt. Sta. Bull. **459**: 47.

Parker, K. G., P. A. Readio, L. J. Tyler, and D. L. Collins. 1941. Transmission of the Dutch elm disease pathogen by *Scolytus multistriatus* and the development of infection. Phytopath. **31**(7): 657–63.

Turner, N., and Nancy Woodruff. 1946. Factors affecting the toxicity of DDT residues. Connecticut Agric. Expt. Sta. Bull. **512**: 93–97.

Whitten, R. R. 1942. Toxic and repellant sprays for the control of elm bark beetles. U. S. Dept. Agric. Circ. **647.**

Whitten, R. R. 1944. Preliminary experiments with DDT in 1944 for the control of the smaller European elm bark beetle. U. S. Dept. Agric. Bur. Ent. and Pl. Quar. **E-670.**

Damage to Corn by the Southern Green Stink Bug[1]

DAN F. CLOWER, *Louisiana Agricultural Experiment Station, Baton Rouge*

ABSTRACT

Adult southern green stink bugs (*Nezara viridula* (L.)) were found severely injuring corn during the summer of 1957. Young corn attacked by the insect showed degrees of injury varying from slight wilting of the central leaves of the whorl to death of the entire plant. On older corn, the ears became distorted and the grain failed to develop. Stink bugs were confined on both young corn and on the developing ears of older corn in an effort to determine the effects of attacks by known levels of infestation. Severe wilting occurred when four pairs were confined on young corn for only 2 days. Confinement of two or more pairs of insects for 13 days resulted in high plant mortality. No deformed ears occurred until the infestation level reached two pairs per ear.

Adult southern green stink bugs, *Nezara viridula* (L.), were found severely injuring corn during the summer of 1957. The infested corn was being grown for hybrid seed in the vicinity of Monroe, Louisiana. Growing corn of all sizes was subject to attack but the greatest damage was observed on young plants 10 to 20 inches in height and to developing ears on older plants.

Infested young corn showed degrees of injury varying from slight wilting of the central leaves of the whorl to death of the entire plant (fig. 1). Extensive feeding punctures were found at the bases of the stalks with dark necrotic areas present within the leaf sheaths and stalks directly beneath these punctures (fig. 2). The terminal buds were frequently dead and in a state of partial decay (fig. 3).

Stink bugs were found at the bases of young plants most frequently when afforded the protection of weeds. They were also observed beneath clods of soil and between the leaves of corn plants.

Suckers subsequently developed from the bases of many of the injured plants after the stink bugs were controlled with an application of one-half pound of dieldrin per acre. Some corn was produced on these suckers but the maturity was delayed about 2 weeks, hence interfering with the practices required in the production of hybrid seed corn.

The insects were observed feeding on the ears and at the nodes over the entire length of the older corn plants. Feeding punctures on the ears frequently resulted in distorted ears and failure of the grain to develop (fig. 4). Little visible injury was found associated with feeding punctures on the stalk.

There was no available information that would indicate the level of infestation which would give economic damage or justify insecticidal control. An effort was made to obtain more information in this regard by caging various numbers of these insects on young corn 10 to 20 inches in height and on the developing ears of older corn.

Adult stink bugs were collected in the infested fields and transferred to Baton Rouge for this work. In test No. 1, the insects were caged on Golden Bantam sweet corn 12 to 15 inches in height. The insects were confined within sleeve cages placed around the bases of the individual corn stalks. The cages were cylinders 8 inches in height and 4 inches in diameter fashioned of 16 mesh galvanized screen. A 12-inch garden stake was attached to one side of the cage as an aid in support. A cloth sleeve was attached to the upper end to facilitate sealing the cage around the corn stalk.

[1] Accepted for publication January 27, 1958.

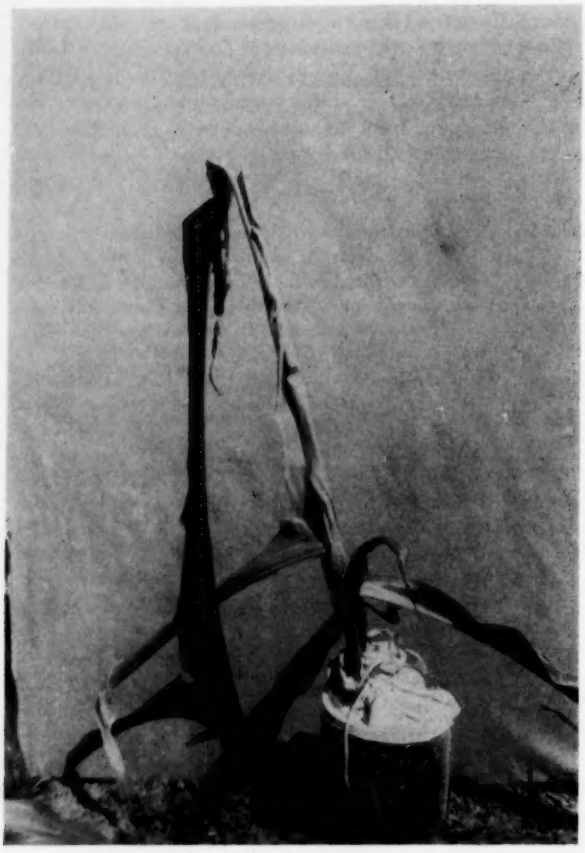


FIG. 1.—Corn plant which has been severely injured by stink bugs.

It had been observed previously that the stink bugs of naturally occurring infestations were most commonly found in pairs at the bases of plants hiding under clods of soil, weeds and other debris. For this reason the sex of all the insects used was determined and a known number of pairs of stink bugs were placed in each cage with a small quantity of grass. Each treatment was replicated six times in a randomized block design and each replicate consisted of a single plant. An index system was utilized to evaluate the resulting injury (table 1).

Test No. 2 was conducted in a similar manner on older corn (var. La. 521) having developing ears present. In this test an individual ear, the node at the base of the ear and the adjacent portion of the stalk were enclosed in 16-mesh galvanized wire screen cages equipped with a cloth sleeve at each end. Ears were selected for uniformity of size and having silks approximately 1 inch in length. These ears were subjected to attack for 10 days. The cages were then removed and the ears examined for evidence of injury (table 2).

Table 1.—Evaluation of injury[a] to Golden Cross Bantam sweet corn subjected to attack by known levels of southern green stink bug infestation.

Replication	Days After Introduction							
	2				13			
	1 Pair	2 Pairs	4 Pairs	None	1 Pair	2 Pairs	4 Pairs	None
I	1	3	3	0	—[b]	4	4	0
II	0	0	3	0	1	3	4	0
III	2	1	3	0	3	2	3	0
IV	0	3	3	0	0	4	4	0
V	1	1	3	0	2	4	4	0
VI	1	1	3	0	1	3	4	0
Average	0.8	1.5	3	0	1.4	3.3	3.8	0

[a] Index—0, no wilting; 1, slight wilting; 2, moderate wilting; 3, severe wilting; 4, dead.
[b] Plant destroyed accidentally.

Table 2.—Evaluation of injury to ears of Louisiana 521 subjected to attack by known levels of southern green stink bug infestation for 10 days.[a]

Treatments	No. of Ears Injured as Indicated			
Pairs per Ear	Nodes with Feeding Punctures	Shucks with Feeding Punctures	Punctures Penetrating to Ear	Deformed Ears
None	0	0	0	0
1	5	5	2	0
2	5	5	4	1
4	5	5	5	2

[a] Five ears per treatment.

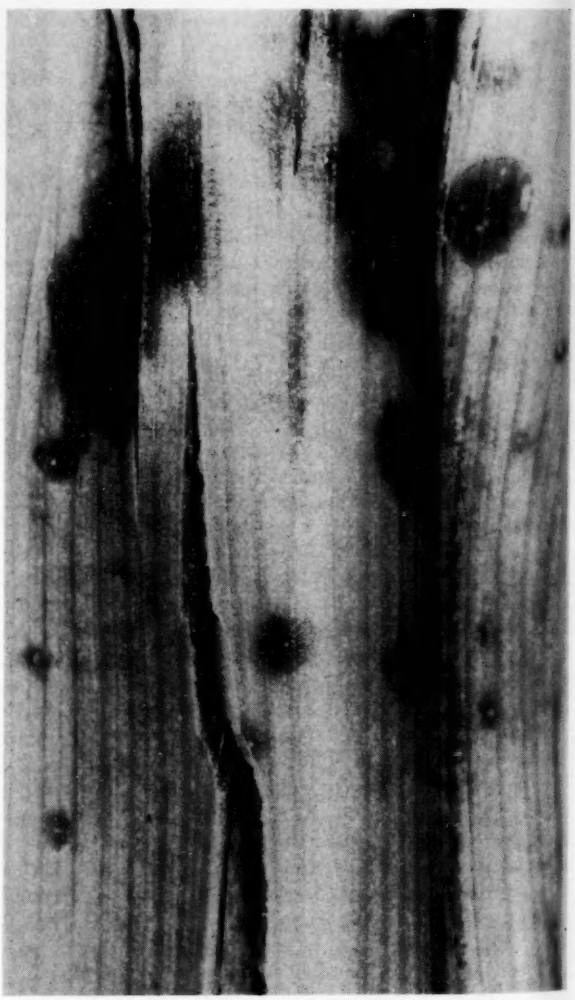


FIG. 2.—The interior of a corn leaf sheath showing stink bug feeding punctures surrounded by dark, discolored areas.

FIG. 3.—Longitudinal section of a corn plant in which the terminal bud was killed showing an area in a state of partial decay within the stalk.

The following treatments were included in both tests:

A = Check
B = 1 pair per cage
C = 2 pairs per cage
D = 4 pairs per cage

RESULTS.—The rapidity with which the symptoms of stink bug injury appeared can be seen by the data presented in table 1. Severe wilting occurred in every case where four pairs of stink bugs were confined on the base of the plants for 2 days. Two pairs caused severe wilting in two of six replications. Confinement of two or more pairs of the insects for 13 days resulted in high plant mortality. The cages were removed at that time and it was found that many of the insects were dead. No additional plants died subsequent to removal of the cage.

FIG. 4.—Distortion of an ear resulting from stink bug feeding.

The corn utilized in this test was growing on land that had been used for testing of insecticides for many years. It is possible that insecticide residue in the soil resulted in some mortality of the insects; hence, the plant mortality may have been less than that occurring under normal field conditions.

Data presented in table 2 show that the caged stink bugs fed readily on the nodes and the shucks, and that the feeding punctures on the shuck frequently penetrated the ear. The grain failed to develop in the areas immediately adjacent to feeding punctures on the ear. Under the conditions of this experiment, no deformed ears occurred until the infestation level reached two pairs. Care was taken when setting up this experiment to select ears of uniform size and maturity. However, when the final examination was made, all ears which had been deformed as a result of stink bug feeding were smaller in size and more immature than the others included in this test. This would indicate the possibility of a stunting effect caused by the feeding.

The Role of Weather and Entomophagous Species in the Natural Control of Insect Populations[1,2]

PAUL DEBACH, *University of California, Citrus Experiment Station, Riverside*

ABSTRACT

Weather sets the basic framework within which certain average but fluctuating insect population densities may be obtained in the absence of other possible regulatory factors. These average population densities may be very high or very low. Weather regulates insect populations only by being of sufficient severity to restrict the size, quality (including food and favorability), and/or numbers of inhabitable spots in a given area—that is, its effect is obtained through interaction with microenvironments. If overlying weather severity in a given spot or area is not relatively great, then, theoretically, populations will be able to increase indefinitely over a period of time unless limited by other factors. Illustrations of population regulation by weather and methods of proof of such regulation are discussed.

The other principal factors capable of regulating insect population densities are (1) natural enemies (parasites, predators, and pathogens), (2) food (quantity and quality), (3) interspecific competition (other than natural enemies), and (4) intraspecific competition. The last three factors usually do not control populations at low (uneconomic) levels and hence are not generally important factors in applied insect ecology.

Natural enemies may regulate insect populations at low levels even if all other factors are exceptionally conducive to increase. Several experimental methods for proving the extent of regulation of insect populations by entomophagous species are given. Thirteen examples of adequate proof of control by natural enemies are listed. A detailed case history of proof of low density population regulation of the California red scale by its natural enemies is given. It is shown that "upsets" of natural enemy effectiveness by adverse factors can result in various differing average population densities of the California red scale (*Aonidiella aurantii* (Mask.), even on adjacent citrus trees.

Throughout the world, entomologists are expending considerable effort in attempts to regulate insect populations by the use of natural enemies. Most scientists familiar with such work agree that in some cases introduced natural enemies have regulated host populations at average densities much lower than those which existed previously. Many also recognize that indigenous natural enemies are responsible for the regulation of host populations at levels much lower than would occur if such natural enemies were absent. When an average insect population density changes markedly to a new average density following either the introduction or the removal of a natural enemy, most people would conclude that the introduction or removal of the natural enemy was responsible for the change, just as they would conclude that a population maintained at a low level year after year in a sprayed plot was due to the insecticide used, particularly if adjacent untreated plots retained high average populations. I believe they would draw the same conclusion without having check plots if the insecticidal treatment resulted in the maintenance of low populations over a period of years, where populations had previously been high over a period of years.

Not all scientists share this viewpoint, and one at the Tenth International Congress of Entomology at Montreal, Canada, in 1956, was quoted in the Montreal Press, as follows: "Dr. Alec Milne, of Newcastle-on-Tyne, said that 'no proof of biological control by parasites' is to be had, and there is 'abundant proof that man has not been able to control insects that way.'" The author and others who heard Dr. Milne's paper agreed that this was essentially what he said. A paper based on this talk at Montreal (Milne 1957) included the following statements regarding the effectiveness of natural enemies: "Biological control work has *not* furnished incontrovertible evidence that any one species can *alone* control another in Nature. Nor is there any stringent evidence that the concerted action of several species can alone control another species, *i.e.*, that the sum of several imperfections amounts to perfection."

In fairness to Dr. Milne, it should be pointed out that even though these statements seem to deny the possibility of success of biological control, he has some cryptic meanings in the above and other statements which leave the door open a crack. The key is his use of the word *alone*. He apparently feels that since parasites are part of an interacting environment, they *alone* cannot be credited with control. I grant that natural enemies need food, air, and other conditions favorable for existence and that they interact with other factors. However, I maintain that if the addition or subtraction of one factor, such as a parasite species, results in a definite change in average population density, then that one factor may be considered responsible for the change and for the new level. Obviously such statements as those made by Dr. Milne could raise considerable doubt in the mind of a reader unfamiliar with biological control studies or with the general subject of factors affecting regulation of insect populations.

Because Dr. Milne is not completely alone in his views; because various schools of thought exist regarding the relative importance of several factors affecting insect populations, particularly weather as compared to natural enemies; because many differences in opinion are due to lack of understanding of the definitions used by various workers; and because actual formal proof of biological control is difficult to obtain and frequently lacking, this paper is written. It is not intended as a technical discussion of the role of all factors in the regulation of insect populations; too much has already been written in this regard along philosophical and mathematical lines, and the situation is already sufficiently muddled as far as the

[1] Paper No. 1023, University of California Citrus Experiment Station, Riverside, California. Accepted for publication January 27, 1958.

[2] Presented as an invitational paper of Section C at the Fifth Annual Meeting of the Entomological Society of America, held at Memphis, Tennessee, December 2–5, 1957.

general reader is concerned. I hope in this paper to get away from the fine distinctions in definition which have apparently kept some population ecologists from understanding one another, and to present convincing arguments and proof to the effect that weather, *in the practical sense*, and natural enemies in certain cases, do regulate the abundance of certain insects.

Other factors such as (1) food (quantity and quality), (2) interspecific competition (other than natural enemies), and (3) intraspecific competition, may, of course, regulate insect populations. However, these three factors usually do not control populations at low (uneconomic) levels and hence are not generally important in applied insect ecology.

Some definitions of terms are given below so that usage may be clear:

"Population"—a group of individuals of the same species living in a limited and defined universe.

"Population regulation (control)"—the maintenance of a population near a certain average density by one or more regulatory factors.

"Biological control"—the utilization of organisms for the economic control of populations of animals and weeds.

"Natural control"—the regulation of populations by any one or more natural factors, unaided by man.

"Regulatory (control) factor"—any mortality factor whose addition to, subtraction from, or marked change in character within the population complex results in a definite change in the average population density. Such factors cause proportionately greater mortality at high densities than at low densities.

"Random mortality factor"—any factor which causes individuals to die, without relation to the density of population. Such factors may cause fluctuations but cannot regulate average population densities.

The Role of Weather in Regulation of Insect Populations.—Students of biological control find themselves from time to time in the position of stating that natural enemies and not weather have been responsible for an observed reduction in average population density of a certain pest. Because they emphasize the effects of natural enemies, it has sometimes been assumed that they consider weather unimportant or even incapable of reducing and/or maintaining potential pest populations at low levels. This assumption has been further strengthened by the controversy involving so-called density-dependent factors (natural enemies or biotic factors) and density-independent factors (weather, etc.). The efforts of some students of biological control and population dynamics have been so strongly directed toward showing that natural enemies, competition, etc., are density-dependent and hence can control a host because they effect a more than proportionate increase in mortality of the host (pest) as its population density increases, that they have intimated or stated that weather is incapable of similar regulation. In the most technical sense they are correct; however, many ecologists have either disagreed with or misunderstood their definitions, because in a general, more practical sense weather acts as a regulatory factor. The following discussion is intended to bring both viewpoints closer together.

Relative weather severity is the factor which either permits insects to exist in a given zone or excludes them. The extent of zones of existence determines distribution. The fact that an insect population exists at all in a given area shows that the weather of the area is favorable to a certain extent. Otherwise, the species would be excluded. These seem obvious statements; however, one might wonder why, if weather is favorable for the survival of some individuals, it isn't for all. (For purposes of simplification, genetic differences between individuals are not considered.) That is, can weather be favorable for survival of some individuals but produce upper limits to increase of the population? The answer is that weather in itself, if it affects a population uniformly and if at all favorable, cannot limit population increase. For this viewpoint weather must be conceived of as having uniform effects upon a given population, such as would be necessary to measure the effects of weather alone.

If we consider a theoretical case in which weather *alone* is responsible for regulation, then, if highly unfavorable—that is, if severe enough to kill all progeny over the number necessary to maintain a 1:1 parent-progeny ratio, extinction would very likely occur within a limited period. Take, for example, a parthenogenetic insect which produces 100 young. If weather alone is to be responsible for control at an average given density, mortality in each generation must be 99%. Anything less than this will result in increase. For example, 95% mortality would result in a 5-fold increase in each generation. Of course, any increase in the severity of weather capable of increasing mortality to more than 99% would probably produce extinction within a more or less limited period of time. It would be difficult to obtain 99% mortality in each of several years without occasionally obtaining 100% from a factor whose severity has no relation to the percentage of mortality it has caused.

Theoretically, then, it seems evident that weather extremes can cause high mortality in an insect population (95% or more in each generation), and yet that population can go on increasing indefinitely. Obviously, with climate-caused mortality of this degree such an increase does not usually occur in nature. From a practical standpoint, weather in many instances does check increase and regulate density of insect populations; however, the technical point is that it does not do it alone. How, then, does weather operate? It must be as follows.

If weather limits populations much below their potential biotic maximum, it must, in general, be unsuitable over the entire population area. That is, the general overlying weather must be severe enough at times to be exterminatory in nature over the area if it is to keep populations low. In nature, however, extermination usually does not occur, because the effect of weather on insect populations is not uniform. Even in severe weather, certain small sections of the population area are more protected and act as shelters or microhabitats which supply niches of favorable weather. In other words, protected or favorable microenvironments must enable some of the population to survive in an otherwise uninhabitable area. The numbers, availability, and quality of these shelters or niches will then determine the population density existing in an otherwise uninhabitable weather zone. These microenvironments may take the form of shelters, better quality or quantity of food, suitable places for oviposition, or any of many direct or indirect broad modifications or reflections

of the usual overlying climate which makes microconditions more or less suitable for survival.

Thus, in a sense, weather considered alone is either tolerable or intolerable, as Nicholson has put it. Insects live only where weather is tolerable; however, it may be tolerable over thousands of square miles or over only small scattered patches of these same thousands of square miles.

Obviously, if weather severity varies, more or fewer favorable microenvironments will be available at various times or in different places. Thus, if intensity of unfavorable weather reduces the number of shelters and hence the number of insects surviving, weather acts in the manner of a density-dependent factor in that the higher the insect population becomes during favorable periods, the greater, percentagewise, will be the reduction during unfavorable periods.

To sum up, weather may regulate insect populations by being of sufficient severity to restrict the size, quality, and/or numbers of inhabitable spots in a given area. If weather severity in a given spot or area is not relatively great, then populations will be able to increase indefinitely over a period of years unless limited by other factors.

Proof of Natural Control by Weather.—Actual proof that weather is the principal factor responsible for regulation of an insect population at low densities may often be quite difficult to obtain. In general, experimental comparisons must be made in which other factors, such as natural enemies, are excluded or are known to have absolutely no effect in population regulation.

Experimental techniques such as those described below for the measurement of natural enemy effectiveness can similarly be applied to the measurement of the total effect of weather. For instance, an insecticide, nontoxic to the host insect, might be used to eliminate over a period of time whatever natural enemies are present in one plot but not in an adjacent plot. If no unusual population increase occurs in either plot, it can be concluded that natural enemies were of no consequence and that weather therefore was responsible for the originally observed population density. (It would also have to be known that the host plant and/or other necessary requisites were favorable to population increase.)

Cases of Natural Control by Weather.—I have studied two insect species occurring in citrus groves in southern California whose populations in certain areas or at certain times appear to be regulated at low densities by weather interacting with microhabitats. One is the purple scale, *Lepidosaphes beckii* (Newm.) the other is the California red scale parasite *Aphytis lingnanensis* Comp.

For more than 50 years the purple scale has caused damage to orange trees in coastal areas. Parasites or predators have never been observed to have any appreciably reliable controlling effect (a recently introduced species is now becoming an important factor but does not relate to the conclusions drawn here) at low densities and so can be ignored as economically satisfactory regulatory factors in this case.

Lepidosaphes beckii was introduced into California about 1889. By 1912 it occurred in every citrus district in California in which it has since been found. It has not spread since that time even though in past years citrus has been grown more or less contiguously from the more coastal areas of Orange and Los Angeles counties, where purple scale has been abundant, through the intermediate areas of Los Angeles County, where purple scale has been scarce to rare, to the interior areas of San Bernardino and Riverside counties, where purple scale has never become established. There is no question but that purple scale has had abundant opportunities to become established in interior citrus areas. This gradation in distribution of the scale is correlated with differences in climates, ranging from that of coastal areas (mild summers and winters and high humidity) through that of intermediate areas to that of interior areas (extreme summers and winters and low humidity). The numerical abundance of purple scale wherever it has been established has been correlated with the same climatic area differences.

Laboratory tests have shown that combinations of high temperature and low humidity are exceedingly detrimental to *Lepidosaphes beckii*. This species is difficult to culture at constant temperatures above 78° F. or at constant relative humidities below 75%.

In climatic areas favorable to purple scale, there is considerable variation in population density between groves, between parts of groves, and within a single tree. All of these differences appear to result from modification of the overlying weather in certain groves (by generally heavier tree foliation), in certain parts of groves (by adjacent windbreaks which cast shadows and influence light, temperature and humidity), or on certain parts of individual trees (90% of the purple scale population occurs on the north, more frequently shaded parts of the trees).

We have accumulated abundant evidence that populations of the California red scale parasite *Aphytis lingnanensis*, as well as those of *A. chrysomphali* (Mercet), are also regulated by the interaction of weather and microhabitats. Considerable data have been published in this regard (DeBach *et al.* 1955). Additional data are as yet unpublished. Briefly, results show that the low winter and high summer temperatures which occur in interior citrus areas frequently greatly depress populations of *Aphytis*. This may approach the point of general extinction, which certainly does occur with some local populations.

Recent comparisons between caged experimentally controlled, initially identical field populations of *Aphytis* which were affected only by weather conditions (surplus food, etc., being available) showed that populations in a mild coastal area did very much better than those in a more severe interior weather zone. In both areas field test populations, in comparison with laboratory check populations, were greatly depressed over the winter, but survival was many times greater in the milder area. During the summer very substantial population increase occurred in the mild area (about one-half the potential shown by laboratory populations); whereas in the severe interior area, extermination occurred. Since field populations of *Aphytis* do not become completely exterminated in interior areas, it is evident that favorable microhabitats so modify the overlying weather during unfavorable periods that scattered survival pockets enable the species to carry on. The reproductive potential of the parasite under such conditions is so greatly reduced, however, that natural control of its host, the California red scale, amost never occurs in interior areas. On the other hand, in many plots in mild

coastal areas, the parasite has satisfactorily controlled the scale.

The Role of Natural Enemies in Natural Control.—According to Milne (1957), "Competing species and other enemies, singly or in concert, cannot control by themselves because they are imperfectly density dependent in action."

Rather than attempt to analyze the cryptic ideas upon which the above statement is based, I think an understanding of this problem in semantics can be resolved by a comparison between the use of parasites and the use of insecticides. When adequate insecticides are applied at suitable intervals to a pest population, that population can be reduced and maintained over an indefinite period at an average density much lower than would otherwise occur. In a sense this is a density-dependent action in that sprays are generally applied as the population density tends to increase. Most people would agree that the insecticide "alone" or "by itself" was the factor responsible for the maintenance of the population at the low average level obtained. In any event the insecticide was the one factor, the critical factor, responsible for the change in average population density from a high to a low level. In the same sense, *adequate* parasites can be considered the one factor responsible for a change in average host population density from a high to a low level if such a change occurs following their addition to the population complex. If Dr. Milne does not think that insecticides *alone* can be responsible for the control of a pest, we can then understand his thinking that parasites *alone* cannot be responsible either.

As far as parasites being "imperfectly density dependent in action" goes, this simply means that if environmental conditions reduce the effectiveness of a parasite species somewhat below its maximum potential, regulation may then occur at a somewhat higher average host population density. It does not follow that because they are "imperfectly" density-dependent, parasites cannot control by themselves as Milne suggests. "Imperfect density-dependence" merely means that a parasite's effectiveness may vary with the environmental conditions which affect it. In spite of such variation regulation may still occur at low population densities.

Admittedly, adverse environmental conditions can so affect a potentially effective parasite as to result in regulation only at a very high host population density. Another way of visualizing imperfect density-dependence is to take two parasite species, A and B, both of which regulate the host at low average densities, but A's level of control is one host individual per unit, whereas B's level of control is two host individuals per unit. Both are effective parasites even though one is better than the other. Now, if species B is thought of as being a population of species A which has been adversely influenced by environmental conditions so that it only effects regulation at double the original host population level (two instead of one per unit), it is evident that "imperfect density-dependence" does not preclude regulation of host populations but merely illustrates that regulation may occur at different levels. This will be demonstrated by the data which follow.

Too much work on the factors affecting field populations has been done in the office. No accurate explanation of what actually happens in nature to a given population can be obtained except in the field. Laboratory studies can help interpret field observations, but the ultimate answer lies in detailed ecological field observation and experimentation. Even then, conclusions regarding factors responsible for population regulation of a species in one area may not apply at different times or to the same species in another area. The ability to draw generalized conclusions will be the exception rather than the rule. We recognize, for instance, that parasites may regulate a host population within economic density limits in one area but fail to do so in another.

Experimental Methods for Evaluating Natural Enemy Effectiveness.—In biological control projects today, one of our most important problems is that of scientific proof of the effect of the natural enemies on the host population. I have been attempting to obtain such proof with various pests and their natural enemies in the field now for 12 years. I have just about concluded that adequate and generally acceptable proof of the effectiveness of natural enemies in host population regulation usually cannot be obtained by correlation between population changes of the host and parasites and/or predators. In most cases it will be impossible or impractical to obtain sufficient detailed data to draw clear-cut conclusions. Adequate life table data may provide the necessary information but similarly will often be impractical to obtain. Such studies may be of great value for other purposes, however.

Detailed field studies may convince the research worker that a given pest population is regulated (*i.e.*, kept from increasing to a higher level) by a particular natural enemy. His problem is to prove to others that such is the case. I am sure that this can best be done by experimental techniques which eliminate one factor or group of factors (*i.e.*, natural enemies) from one plot and show by comparison with a plot otherwise comparable but having natural enemies present, what difference in an average population density develops. In other words we make a simple experimental comparison involving the addition or subtraction of one factor, essentially as is done in insecticidal field tests. The development of adequate techniques for making such comparisons is not always easy, but several have been found and carried out. These include (1) the insecticidal check method (DeBach 1946), (2) the sleeve-cage check method (DeBach *et al.* 1949), (3) the biological check method (DeBach *et al.* 1951), (4) the manual removal of natural enemies (Fleschner *et al.* 1955), and (5) the insecticidal trap method (unpublished data, Bartlett & DeBach 1957).

The last-named method involves the elimination or retardation of natural enemies by surrounding an untreated area with an area treated with appropriate insecticides. Migrating natural enemies are poisoned as they move through the treated area, and their population in the central untreated area is gradually reduced.

These experimental methods comprise a comparison in time between two initially comparable pest populations in adjacent plots. The only difference, if the tests are properly conceived and executed, between the two populations is that natural enemies are eliminated or depleted and retarded as a mortality factor affecting one of the paired populations. Comparison of differential population growth in the two plots, one "with" and one "with-

out" natural enemies, gives a striking picture when effective natural enemies are involved. Often it is desirable to carry out two or more of these methods in the same grove or population area. Between them it is usually possible to rule out or evaluate other possible control factors such as weather or host-plant resistance.

In spite of the presentation of this type of proof by myself and other authors, Milne (1957, pp. 204–5) remains unconvinced. He questions the sampling techniques which were statistically reliable even though, originally, details were not published; he quoted but completely misunderstood one very important table which showed that DDT as used in the "insecticidal check method" did not in itself promote scale increase because no significant differences in scale populations occurred between treated and untreated lemon branches. No parasites were involved in this test, as was clearly stated, but Milne missed this and, apparently believing that parasites were involved, decided that because there were no differences between treatments (the author's very conclusions), the conclusions were unjustified. He then ascribes to "the Riverside School" (quoting from papers by DeBach and coauthors) the belief "that enemies are responsible for controlling citrus pests wherever natural control occurs." This statement was never made by our group and is merely a mental extrapolation by Milne from the facts we presented. We have always recognized that factors other than natural enemies may be responsible for natural control at low population densities. Another unwarranted statement was that "they never mention bird predation." Neither did we mention any of dozens of other theoretically possible causes of mortality which were of no consequence.

Because there still seems to be some question as to the adequacy or understanding of previous work involving *proof* of natural and/or biological control, I am going to rediscuss and elaborate upon one particular case—that of the California red scale, *Aonidiella aurantii* (Mask.), and its control by natural enemies. This study illustrates many facets of population dynamics.

Similar experimental *proof* of cases of biological or natural control also has been developed with several other pests, including the following: on citrus: the yellow scale (*Aonidiella citrina* (Coq.)) (DeBach 1955), citrus mealybugs (DeBach *et al.* 1953, p. 247; DeBach 1946, p. 696), soft scale (*Coccus hesperidum* L.) (DeBach 1951, p. 765), aphids (DeBach 1951, p. 764), citrus red mite (*Panonychus citri* McG.) (DeBach *et al.* 1950, p. 810; DeBach 1951, p. 765; Fleschner 1952), and the cottony-cushion scale (*Icerya purchasi* Mask.) (DeBach 1946, p. 696; DeBach & Bartlett 1951, p. 375); on avocados: the omnivorous looper, the six-spotted mite (*Eotetranychus sexmaculatus* (Riley)), the long-tailed mealybug (*Pseudococcus adonidum* (L.)), the avocado brown mite, and the latania scale (Fleschner *et al.* 1955); on strawberries: the cyclamen mite (*Steneotarsonemus pallidus* (Banks)) (Huffaker & Spitzer 1951, Huffaker & Kennett 1956); and on apple trees: phytophagous mites (Lord 1956).

Methods of Sampling California Red Scale.—For 10 years some six of us have been engaged principally in detailed field and laboratory studies of the ecology of the California red scale and its parasites and predators in untreated citrus groves (no insecticides) throughout southern California. More than 20 groves have been studied critically during this period, the work involving among other things detailed biweekly or monthly partial census counts of the scale and its natural enemies. The shortest study of this type in a particular grove covered a period of 3 years, the longest has now covered 10 years. Many other short-term studies have been carried out in these and other groves. Invaluable data on population dynamics have been accumulated, only a small portion of which can be presented here as illustrative material. Some results of these studies have been published (DeBach 1951, 1955, DeBach *et al.* 1950, 1953, 1955); others remain to be published.

A discussion of the methods used for sampling the California red scale and its parasites has appeared in an earlier publication (DeBach *et al.* 1950, pp. 785–7). Because we are not trying in the present paper to correlate natural enemy populations with those of the host in order to demonstrate natural enemy effectiveness but are trying rather to show what changes in scale populations occur when one factor, that of natural enemies, is eliminated, only the adequacy of the California red scale population sampling method is of consequence here. Suffice it to say that detailed census counts are available on both predator and parasite populations.

It was found early in these studies that in an untreated citrus grove in which natural control occurred at low densities, the scale populations on individual trees were remarkably uniform. (Ant infestations can result in variable red scale populations between trees; these or other "upsets" are recognized and taken into account when necessary.) Hence, one or a few trees usually represent an adequate and authentic picture of the grove as a whole. We have found that as the population on one tree changes, in time, so do those on the others. Moreover, one tree in a grove can be considered a "universe" and studied as an entity. This also holds true for isolated trees, except that natural enemy fluctuations on such trees may be greater, owing to the occurrence of more emigration than immigration, and natural control may not be quite so effective. Since there is virtually no movement of California red scales to or from a given tree, the red scale population of a given tree may be considered peculiar to that tree alone and relatively uninfluenced by adjacent trees.

As a general rule, five permanent census trees were selected in a given grove and sampled regularly thereafter. An average of these five trees gives a very reliable sample for the grove as a whole. We have always found that time trends on any one of the five trees represent rather closely the trends on the other trees. Hence, an accurate sample based on one tree is the key to the entire sampling problem. The fact that frequent regular periodic samples are taken makes each sample a check on previous and subsequent samples and thus adds additional weight to the validity of any given sample.

In the early studies we sampled leaves, fruit, and twigs, but later we determined statistically that a twig sample for scale population density was a highly reproducible index of the population. This sample consisted of counting the number of live red scales on 3-inch sections of 24 twigs of medium-aged growth. Six twig units were drawn from

each of four quadrants of the tree. In each quadrant, twig units were selected from high, medium, and low levels, and at each level one unit was selected inside and one at the periphery of the tree. Actual choice of twigs was made at random.

The total number of live scales on such a sample gives an accurate index to the population on the tree as a whole. It is highly reproducible on replicate counts and by different counters. The statistical reliability of the California red scale sample is shown in table 1 for different sample population densities.

From the data of table 1 it is evident that this sampling method gives a very accurate population density index for any given tree, with rather small expected variation about the mean, as judged by the limits at the 0.05 level of probability. Population estimates are extremely close at the lower population ranges of 0 to 5, 6 to 10, and 11 to 20 scales per individual tree sample. Somewhat greater numerical variation between samples can be expected in the high population density categories, but in this case the population density is so high as to minimize the importance of this variation. The limits of variation about the mean at the 0.05 level of probability can be used for interpretation of the significance between the different population density ratings given later in table 2 and figure 1.

Table 1.—Statistical reliability of California red scale population sample.[a]

No. of Trees Counted[b]	Sample Population Density Ratings per Tree: Range	Mean	Limits at 0.05 Level of Probability: Lower	Upper	Qualitative Rating of Scale Population Density	
21	0–5	0.76	0.23	1.39	rare	Commercial control
5	6–10	7.03	5.68	8.59	scarce	Commercial control
12	11–20	13.97	10.06	18.46	light	Commercial control
3	21–30	25.45	22.92	28.12	medium	No commercial control
5	31–50	38.37	31.72	45.64	heavy	No commercial control
2	51–100	67.18	56.62	78.63	very heavy	No commercial control
2	101–200	131.78	110.79	154.58	very heavy	No commercial control

[a] Statistical analysis by Dr. M. J. Garber, Assistant Biometrician, University of California Citrus Experiment Station, Riverside.
[b] Five replicate counts per tree.

Population Dynamics of the California Red Scale.—As a background for the detailed discussion which follows, a general statement of our carefully considered conclusions regarding the population dynamics of the California red scale on citrus in southern California may help to orient the reader.

The California red scale is the worst pest of citrus in California and, in the absence of effective parasites, major annual or semiannual insecticidal treatments are always required. Weather has no effect of any practical consequence on the California red scale on citrus in the major California citrus areas extending from San Diego to Fresno, as far as the pest status or average population density of the scale is concerned. Relative weather severity affects the rate of increase of the scale to an extent (relative rate of increase may be of importance in groves regularly treated with insecticides), but controlled tests throughout the citrus areas have shown that light scale infestations will rapidly increase manyfold to the extent of tree defoliation within 1 or at most 2 years if natural enemies are not present and insecticides are not applied. Hence, weather is predominantly favorable for scale increase and will permit relatively rapid increase to the limit of the food supply.

Host-plant resistance to scale increase is not a regulatory factor except with mandarin type, loose-skinned oranges. California red scale fails to reproduce to any extent on these varieties; however, they represent an inconsequential percentage of the citrus acreage. Other citrus varieties may affect the relative rate of increase of red scale as does weather, but again controlled tests on all commercial varieties have shown that light scale infestations will increase to the point of completely defoliating the tree within the short period of 1 to 2 years, if natural enemies or insecticides are not involved. Red scale populations tend to increase most rapidly on lemons and grapefruit and less radidly on navel and Valencia oranges.

Variations in edaphic factors have not been observed to have any appreciable effect on California red scale populations. It follows, therefore, that control of red scale in the citrus areas of California must result either from the use of insecticides or from the activity of natural enemies. From the time of the introduction of red scale into California in the 1870's until recently, it has generally been assumed that natural enemies play little if any part in economic regulation of red scale populations. Occasionally, *Aphytis chrysomphali* (Mercet), the tiny golden-chalcid parasite, or various predators were suggested as being of some importance, but no ecological studies were ever carried out to evaluate their importance.

In 1948 we began ecological studies of the California red scale in various groves throughout the citrus areas of southern California. Only groves which had been untreated with insecticides for several to many years were chosen, so that the possible adverse effect of chemical treatment on natural enemies could be ruled out as a factor. Such untreated groves were very difficult to find and represented only a very small fraction of the total citrus acreage. In some of these study plots the red scale was heavy, but in others, surprisingly, it was very light. In the lightly infested groves virtually only one species of natural enemy was found to be of any consequence, the parasitic golden chalcid *Aphytis chrysomphali*. In medium to heavy red scale infestations, various predators (*Chilocorus* spp., *Lindorus lophanthae* (Blaisd.), and others) may become common, but they have never been known to maintain a red scale population at a low average density.

Eventually, in many low density plots we were able to prove by controlled tests involving the exclusion, elimination, or retardation of *Aphytis* that scale populations would increase rapidly to the limits of the food supply in the near absence of this parasite. These tests may also exclude or depress other natural enemies which normally operate at high red scale population densities. It is recognized that if *Aphytis* were not present but predators or other parasites were, the others usually would stop scale increase somewhere below the limits the tree can tolerate but that extensive tree damage would occur and the economic threshold would be greatly exceeded. Hence, *Aphytis* was the principal factor responsible for regulation of red scale populations at low densities.

Such low density regulation was not evident in all

groves or areas, however, and eventually it was found that two major adverse factors could negate the potential effectiveness of the parasites. *Aphytis* was so adversely affected by unfavorable weather conditions in some areas that regulation of red scale at low population densities did not occur. Ants interfering with or killing *Aphytis* adults also resulted in failure of the parasites to regulate the scale populations satisfactorily at low levels.

In coastal zones such as Orange County, satisfactory natural control of red scale by *Aphytis* occurred in all untreated groves studied if ants were not involved or were eliminated. In slightly intermediate climatic zones such as the Fallbrook-Escondido area of San Diego County, excellent natural control also occurred. In truly intermediate climatic zones such as inland Los Angeles County, regulation was satisfactory in some plots but not in others. In interior climatic zones such as the Riverside-San Bernardino area or the San Fernando Valley, satisfactory population regulation of red scale by *Aphytis* at low densities virtually never occurred. The critical factor adversely affecting *Aphytis* in the last-named zones was weather, not ants (DeBach *et al.* 1955). From the practical standpoint it has been demonstrated that biological control of California red scale, as well as of other orange tree pests, can be obtained on several thousand acres of citrus in San Diego County. Previously, annual insecticidal treatment was thought to be necessary (DeBach 1952).

A Case History—The Sullivan Lemon Grove, Santa Barbara.—The study in the Sullivan lemon grove will serve as a typical example of similar studies in other groves which led to the discovery that natural enemies do control California red scale populations at low average densities in certain groves. This was one of the two groves found in Santa Barbara County in 1948 which the owners agreed to leave untreated with insecticides. The original objective was to study the ecology of the natural enemies present with the idea of perhaps supplementing their supposed weaknesses through periodic colonization of laboratory-reared individuals. Predators were at first thought to offer the most promise. *Aphytis chrysomphali* was known to be present but was thought to be ineffective. It was not anticipated that the natural enemies already established were capable, unaided, of reducing the original medium-to-heavy scale infestation to low average densities.

When first observed in 1948 this grove was not commercially clean of red scale. The infestation was not causing serious tree damage, but occasional twigs were heavily enough infested to be killed. The Argentine ant, *Iridomyrmex humilis* Mayr., was abundant on the trees and was obtaining honeydew from the citrus mealybug, *Pseudococcus citri* (Risso), and the soft (brown) scale, *Coccus hesperidum* L. At that time we did not suspect the Argentine ant of being indirectly responsible for influencing red scale population increase, because the ant pays no attention to the red scale.

Ten census trees were established in April 1948, and counts were made at monthly intervals thereafter on each tree until June 1953, a period of a little more than 5 years. The census sampling methods used were those described above.

Biological Check Tests.—In July 1948, ants were eliminated from 6 of the 10 census trees, so thereafter two different plots were studied, one with ants and one without. By the end of 1948 the differences in California red scale populations in these plots made it readily evident that in some way ants were benefiting the red scale. Subsequently, this was borne out in many other groves in different areas over a period of several years, and it was found that the ants were killing or disturbing the natural enemies of the red scale. This offered a method of evaluating the effectiveness of the natural enemies, even though their efficiency was merely decreased, not completely eliminated (DeBach *et al.* 1951).

Sleeve-Cage Check Tests.—In April 1948, paired sleeve-cage tests were set up over branches on three different trees; in July two more tests were set up on two other trees. The initial red scale populations involved were light to medium. Ants were eliminated from these trees. Each tree had one closed, DDT-impregnated organdie cloth sleeve and one untreated sleeve open at the ends. The closed, DDT-treated sleeves completely eliminated natural enemies inside but had no effect on the sessile red scale. The open, untreated sleeves permitted natural enemies to work normally as they did on the rest of the tree. Biological tests showed micrometeorological conditions to be similar within the open and closed sleeves and also showed that the "climate" within the sleeves had little if any favorable effect as compared with red scale survival and increase outside the sleeves. A comparison of scale populations within the open and closed sleeves and on the whole tree therefore showed the total effectiveness of the natural enemies in retarding and regulating potential red scale increase (DeBach *et al.* 1949).

Counts in this case were not necessary. Of the three original light-to-medium scale-infested branches in the DDT-treated sleeves freed in April from the mortality caused by natural enemies, one had such rapid scale population increase that complete defoliation occurred by December (8 months later); another had some twigs dying from incrusted scale by December but lingered on with considerable defoliation and a heavily incrusted scale population until the death of the whole branch the following August (16 months later); the third sleeve-covered branch showed heavy scale infestation by December and kill of half of the twigs by the following October (18 months later). During this same period, April 1948 to October 1949, scale populations in the open untreated sleeves and on the rest of the tree decreased. This could only have been due to the action of natural enemies, because climate and host plants were demonstrated to be highly favorable.

These sleeve-cage checks prove that natural enemies were repsonsible for satisfactory natural control of red scale on these trees, and that without their action red scale could have increased rapidly to the point of complete tree defoliation within 8 months, and to the point of complete tree destruction within 16 months. Similar results were obtained repeatedly in other experiments in other areas.

Insecticidal Check Tests.—A DDT-treated insecticidal check tree was established in February 1949. Subsequent light applications of DDT at monthly intervals effectively reduced, but did not completely eliminate, natural enemy activity. DDT, as used, had no appreciable toxic

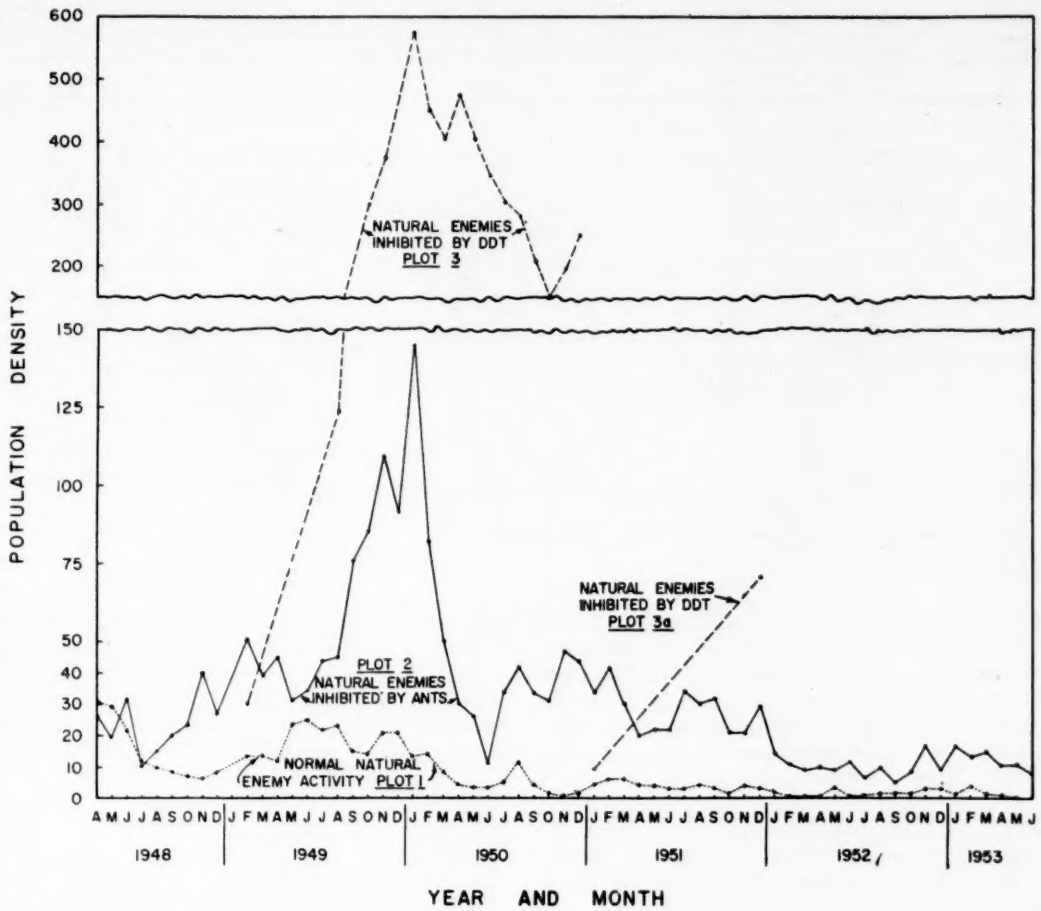

600
500
400
300
200
150
125
100
75
50
40
30
20
10
0
POPULATION DENSITY
NATURAL ENEMIES INHIBITED BY DDT PLOT 3
NATURAL ENEMIES INHIBITED BY DDT PLOT 3a
PLOT 2 NATURAL ENEMIES INHIBITED BY ANTS
NORMAL NATURAL ENEMY ACTIVITY PLOT 1
A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J
1948
1949
1950
1951
1952
1953
YEAR AND MONTH

Fig. 1.—Different degrees of natural control as demonstrated by population density trends of the California red scale on lemon trees in adjacent plots under conditions of experimentally varied natural enemy effectiveness: (Plot 1) excellent natural control under conditions of normal natural enemy effectiveness; (Plot 2) fair natural control with natural enemy activity retarded by ant interference; (Plots 3 and 3a) very poor natural control with natural enemy activity greatly retarded by the toxic effects of DDT sprays. Neither ant interference nor DDT residue had any appreciable effect on red scale populations except indirectly through adverse effects on natural enemies.

effect on the red scale and did not modify the tree physiologically to favor red scale survival and increase (DeBach 1955). As a result of natural enemy retardation by DDT, red scale populations increased very rapidly to a high density. The effectiveness of natural enemies was thus demonstrated by this third experimental method. A second DDT check tree replaced the first in January 1951, because of extensive scale-caused damage to the original tree (table 2 and figure 1).

Census Data Population Trends.—The census data obtained from the California red scale population samples are tabulated in table 2 for the DDT insecticidal check tree tests, the ant-biological check trees, and the ant-free natural control trees. Chronological trends based on these data are shown in figure 1.

The most obvious and perhaps the most significant results shown in table 2 and figure 1 are that red scale populations can assume strikingly different densities in adjacent climatically identical "universes" (trees). With red scale, as stated above, one tree can be considered a "universe" because the scales are predominantly sessile and movement between trees is negligible. Here, with the same potential for scale population increase on each tree, three distinctly different population trends developed in different plots. The only variable between plots was natural enemy activity. Obviously, therefore, there is no such thing as one average or normal population density equilibrium position for a given species except under one particular definite set of environmental conditions.

Inasmuch as the three greatly different population-density trends are due merely to the effect of different adverse factors on natural enemy effectiveness, it is evident that Milne's (1957) statement that natural enemies cannot control by themselves because they are imperfectly density-dependent does not hold. Even in the proven cases of excellent natural control at very low scale population densities, the natural enemies are not operating in a perfectly density-dependent manner because winter cold seriously reduces their numbers and efficiency (DeBach *et al.* 1955). That is, they could do better under ideal conditions. However, assuming for purposes of argument that they were perfectly density-dependent in this case,

Table 2.—Average population density time trends of California red scale per tree sample in various plots: (Plot 1) normal natural enemy activity (ant-free trees), (Plot 2) partially depressed natural enemy activity (ant-infested trees), and (Plot 3) strongly depressed natural enemy activity (DDT-treated trees) in Sullivan lemon grove, Santa Barbara, California (1948-1953).[a]

	1949		1949			1950			1951			1952		1953	
	Plot. No.		Plot No.			Plot No.			Plot No.			Plot No.		Plot No.	
Month	1 Ant-Free Trees	2 Ant-Infested Trees	1 Ant-Free Trees	2 Ant-Infested Trees	3 DDT-Treated Tree	1 Ant-Free Trees	2 Ant-Infested Trees	3 DDT-Treated Trees	1 Ant-Free Trees	2 Ant-Infested Trees	3 DDT-Treated Tree	1 Ant-Free Trees	2 Ant-Infested Trees	1 Ant-Free Trees	2 Ant-Infested Trees
January	Plots started		—	No count	—	13	145	575	4	33	9[d]	2	14	2	17
February	in		13	51	30[b]	14	82	450	6	42	—	1	11	4	13
March	April		13	39	—	8	50	410	6	30	—	1	9	2	15
April	31	26	12	45	—	4	30	475	4	20	—	1	10	1	11
May	29	19	23	31	—	3	26	410	4	22	—	3	9	0	11
June	22	32	25	34	—	3	12	340	3	22	—	1	12	0	8
July	12	10	22	43	—	5	33	310	3	34	—	1	7	Termination of study	
August	10	15	23	45	123	12	42	280	4	30	—	2	10		
September	8	20	15	76	—	4	33	220	3	32	—	2	5		
October	7	23	14	85	300	2	31	157	2	21	—	2	8		
November	6	40	21	109	375	1	47	195	4	21	—	3	17		
December	8	27	21	92	—	2	43	250[c]	3	29	71[e]	3	9		

[a] No pest control treatment applied after 1947. Sample count densities below 5 represent very light scale populations; 5 to 10, light scale populations and good economic natural control; 11 to 15, light to medium scale populations and a borderline economic infestation.
[b] Initial rating and spray of first DDT-treated tree.
[c] Terminated in December 1950. New DDT-treated tree started in January 1951.
[d] Initial rating and spray of second DDT-treated tree.
[e] Terminated in December 1951.

then when ant interference was involved they became imperfectly density-dependent and yet still exerted population regulation at a somewhat higher but still fairly low density as compared to the potential red scale population density that would develop if natural enemies were made more imperfectly density-dependent by the adverse effect of DDT. Thus density-dependence is seen to be a matter of degree which can result in populations being regulated at any of many possible average densities.

Natural Control under "Ideal" Conditions.—Where there were no known factors except weather which might be adverse to the natural enemies (fig. 1, plot 1), a low red scale population was maintained from the time ant interference was eliminated in July 1948 until the termination of the tests in June 1953. Actually a slow downward trend is evident over the entire 5-year period, which terminated in extremely low red scale populations. Any rating below 10 represents a very light red scale infestation and very satisfactory control from a commercial standpoint.

At the beginning of the tests on these particular trees, when ants were eliminated in July 1948, the red scale infestation bordered on a light-medium commercial rating (dangerous only from the standpoint of potential increase). A slight downward trend developed during the last half of 1948; then, following the coldest spell of the entire 5-year study, in January of 1949, when snow and ice covered the trees and natural enemies were depleted out of all proportion to the more tolerant red scale, an upward trend occurred through the summer of 1949 which was reversed as natural enemies again increased in numbers in midsummer and fall. This 1949 population on the "natural control" trees was the highest of the entire 5-year study, but from the commercial standpoint this would mean, at most, little loss from fruit cullage due to presence of scales. Such a population is far below the level at which tree damage occurs.

According to our sampling method, a rating of "20" means 20 live scales present on a standard sample of 24 three-inch sections of twig, or less than one scale on every 3 inches of twig. Such a population would not be readily evident to the casual observer. In 1950 the population density decreased to an extremely low status, which continued through 1951 and 1952 and to the termination of the study in 1953. That natural enemies were responsible for this excellent natural control is evident by comparison with the other, much higher population trends on adjacent trees (see "Natural enemies inhibited by ants" and "Natural enemies inhibited by DDT," fig. 1), where the effectiveness of natural enemies was greatly reduced by the presence of ants or DDT.

Natural Control under Conditions of Ant Interference.—The trees in this plot were adjacent to and similar to the trees in the plot described above, except that ants were permitted to remain in this case and, as has been explained previously, to interfere with natural enemies (see curve labeled "Natural enemies inhibited by ants," fig. 1). From the beginning of the study in April 1948, until July, both groups of trees (plots 1 and 2) were ant-infested, and population densities were roughly comparable. When ants were eliminated, in July, from the trees of plot 1, average population densities on the two groups of trees were very close. The group of trees with ants showed marked increase in scale populations from July 1948 to February 1950, whereas the group without ants remained with relatively light scale populations. The striking increase of scale on the ant-infested trees during 1949 was undoubtedly due to the ants further inhibiting the few natural enemies surviving the severe freeze in January 1949. Throughout the 5-year period of comparison, the red scale populations were always appreciably higher on the ant-infested trees than on the ant-free natural control trees. For most of this period, natural control on the ant-infested trees occurred at levels above the threshold of economic damage; this is nonetheless an example of natural control and, if comparison is made with the potential for increase of the scale as partially demonstrated by that occurring on the DDT trees, is not inconsiderable natural control if the economics of the crop are not considered.

The fact that ants have no effect on red scale, except in-

directly through inhibition of natural enemies, proves that the observed low scale population on natural control trees without ants is due to the action of natural enemies alone. The ant and DDT check tests eliminate the possibility that climate might be acting in a regulatory manner.

Natural Control under Conditions of Insecticidal Interference.—DDT sprayed periodically on a tree in very light dosages is toxic to natural enemies of the California red scale but has little toxic effect on the red scale except perhaps to kill some active "crawlers" (motile, newly born scales). It will be recalled also that DDT does not stimulate red scale increase through some hypothetical direct or indirect physiological or nutritional effects. Such use of DDT results (when effective natural enemies are present originally) in a purposely induced insecticidal "upset" similar to the numerous "upsets" in natural balance reported for many insect species on many crops during the past few years. Obviously, if scale populations increase markedly to new levels following the use of a differential insecticide whose only effect is to retard natural enemies, the climate and host plant must be eminently suitable. This is demonstrated by the terrific red scale population increase which occurred when natural enemy activity was depressed by DDT (see curve labeled "Natural enemies inhibited by DDT," fig. 1, plot 3).

The population increase of California red scale on the DDT-sprayed tree, from its initiation in February 1949 to the end of the year, was a biological explosion. The increase was 20-fold in about 10 months and would have been greater if crowding effects had not depressed the rate of scale increase. Scales were so massed together on much of the twig and fruit surface of the tree by the end of December that the tree was unable to support such a heavy scale population and showed nearly complete defoliation, along with some twig-kill, by January 1950. This, plus (1) intraspecific competition due to crowding, (2) interspecific competition for space and food with mealybugs and red mites which also became abundant, and (3) possibly some winter-caused mortality led to a downward trend in the extremely heavy scale population until March. The April counts showed a tendency for the scale density to increase again, but this trend reversed and continued downward until October and then increased again until the test was terminated in December 1950.

Abandonment of counts on this tree became necessary because excessive damage from red scale made the tree unsuitable for test purposes. The downward trend from April to October 1950 was apparently due to natural enemies, both predators and *Aphytis*, which become abundant in spite of the DDT. Relatively speaking, however, they were not abundant in relation to the scale population. This same occurrence has been noted in many other DDT check tests in other groves over the years. At first, natural enemies seem to be excluded nearly completely, but when scale populations become extremely heavy, predators and *Aphytis* may become quite common numerically and actually reduce the scale population somewhat, but never to a density low enough to prevent extensive tree damage by the scale.

A second DDT insecticidal check comparison (see curve labeled "Natural enemies inhibited by DDT," plot 3a, fig. 1) was started in January 1951. The purpose was merely to replicate the 1949–50 DDT tests and to obtain an indication as to potential rate of increase of the red scale in 1951. This test started at a rather low population density rating of 9 in January and was terminated with a high population density rating of 71 in December. This represents an 8-fold increase for the year, whereas there was no increase whatsoever on the ant-free natural control trees. Comparable or greater increases have been obtained without exception with each DDT-insecticidal check test we have ever run, providing, of course, that adequate natural enemies were present in the grove. Wherever such "explosions" of red scale follow the use of DDT, they provide proof that natural enemies were responsible for the regulation of the former population at the originally observed density.

References Cited

DeBach, Paul. 1946. An insecticidal check method for measuring the efficacy of entomophagous insects. Jour. Econ. Ent. **39**(6): 695–7.

DeBach, Paul. 1951. The necessity for an ecological approach to pest control on citrus in California. Jour. Econ. Ent. **44**(4): 443–7.

DeBach, Paul. 1952. Biological control of red scale in San Diego County. California Citrog. **37**()4: 136–7, 58, 59, 60.

DeBach, Paul. 1955. Validity of the insecticidal check method as a measure of the effectiveness of natural enemies of diaspine scale insects. Jour. Econ. Ent. **48**(5): 584–8.

DeBach, Paul, and B. R. Bartlett. 1951. Effects of insecticides on biological control of insect pests of citrus. Jour. Econ. Ent. **44**(3): 372–83.

DeBach, Paul, E. J. Dietrick, and C. A. Fleschner. 1949. A new technique for evaluating the efficacy of entomophagous insects in the field. Jour. Econ. Ent. **42**(3): 546–7.

DeBach, Paul, E. J. Dietrick, and C. A. Fleschner. 1953. Natural control of the California red scale in untreated citrus orchards in southern California. Proc. Seventh Pac. Sci. Congress, 1949, Vol. **IV**: 236–48.

DeBach, Paul, E. J. Dietrick, C. A. Fleschner, and T. W. Fisher. 1950. Periodic colonization of *Aphytis* for control of the California red scale. Preliminary tests, 1949. Jour. Econ. Ent. **43**(6): 783–802.

DeBach, Paul, T. W. Fisher, and John Landi. 1955. Some effects of meteorological factors on all stages of *Aphytis lingnanensis*, a parasite of the California red scale. Ecology **36**(4): 743–53.

DeBach, Paul, C. A. Fleschner, and E. J. Dietrick. 1950. Studies of the efficacy of natural enemies of citrus red mite in southern California. Jour. Econ. Ent. **43**(6): 807–19.

DeBach, Paul, C. A. Fleschner, and E. J. Dietrick. 1951. A biological check method for evaluating the effectiveness of entomophagous insects. Jour. Econ. Ent. **44**(5): 763–6.

Fleschner, C. A. 1952. Host-plant resistance as a factor influencing population density of citrus red mites on orchard trees. Jour. Econ. Ent. **45**(4): 687–95.

Fleschner, C. A., J. C. Hall, and D. W. Ricker. 1955. Natural balance of mite pests in an avocado grove. California Avocado Soc. Yearbk. **39**: 155–62.

Huffaker, C. B., and C. E. Kennett. 1956. Experimental studies on predation: Predation and cyclamen-mite populations on strawberries in California. Hilgardia **26**(4): 191–222.

Huffaker, C. B., and C. H. Spitzer. 1951. Data on the natural

control of the cyclamen mite on strawberries. Jour. Econ. Ent. **44**(4): 519–22.

Lord, F. T. 1956. The influence of spray programs on the fauna of apple orchards in Nova Scotia. IX. Studies on means of altering predator populations. Canadian Ent. **LXXXVIII**(3): 129–37.

Milne, A. 1957. The natural control of insect populations. Canadian Ent. **LXXXIX**(5): 193–213.

Tests with Compounds Showing Synergistic Action with Rotenone against Body Lice[1]

M. M. COLE and P. H. CLARK,[2] *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*

ABSTRACT

Sulfoxide and piperonyl butoxide were highly synergistic with rotenone in laboratory tests against body lice, *Pediculus humanus humanus* L.; (Anoplura:Pediculidae), and 1,2-methylenedioxy-4-[2-(octylsulfonyl)propyl]benzene, 5-butyl-5-ethyl-2-(3,4-methylenedioxy)-*m*-dioxane, and the *alpha*-allylpiperonyl ester of fencholic acid showed a lower degree of synergism. Rotenone alone was less toxic to lice than the other compounds. A powder containing 0.5% of rotenone and 5% of sulfoxide was highly effective for a longer period than one containing 0.2% of pyrethrins and 2% of sulfoxide in tests against DDT-resistant lice on human subjects.

Rotenone, in ground derris or cube root or their extracts, was used for the control of the body louse, *Pediculus humanus humanus* L., prior to World War II, when it was displaced by DDT. The development of resistance to DDT and other chlorinated hydrocarbons in body lice, as in many other insects, has stimulated the reconsideration of the older insecticides as well as the search for new ones.

Bushland *et al.* (1944) reported that a powder containing 1% of rotenone was highly effective against body lice in practical tests with treated underwear, but caused severe dermatitis in the scrotal region of the research subjects. Trembley (1943) also reported that derris powder used for the control of pubic lice occasionally caused dermatitis. However, rotenone has been found to be highly synergistic with certain other compounds, and such combinations may permit its use at lower concentrations for the control of body lice.

Pearson (1935) reported that pine oil increased the toxicity of derris extract in Peet-Grady tests against house flies. Pierpont (1939) reported that ethylene glycol ether of pinene (D. H. S. Activator) also increased the effectiveness of rotenone against house flies. However, Douglas & Furman (1949) found that neither piperonyl butoxide nor *n*-octyl bicycloheptene dicarboximide (MGK-264) made rotenone more toxic to cattle grubs. Moore (1950) reported that a dip containing piperonyl butoxide and rotenone was very effective against the brown dog tick, and a dust containing piperonyl cyclonene with pyrethrins and rotenone against the American dog tick. However, it is not certain that the effectiveness of rotenone was increased by the combination with these materials in these tests against ticks, for it apparently was not tested alone. Roberts *et al.* (1953) reported that, for the control of cattle grubs, an aerosol containing piperonyl cyclonene with rotenone was unsatisfactory and piperonyl butoxide did not increase the effectiveness of rotenone.

This paper describes some laboratory tests with rotenone in combination with selected compounds, some of which had been shown to be synergistic with organic phosphorus insecticides (Eddy *et al.* 1954a), allethrin (Eddy *et al.* 1954b), cyclethrin (Burden & Cole 1956), or pyrethrins (unpublished data).

METHODS.—The methods used in these studies were essentially those described by Bushland *et al.* (1944) and modified by Eddy *et al.* (1954a). The beaker tests consisted of saturating circular pieces of Army OD wool cloth with acetone solutions of the test materials, allowing them to dry on pins, and then placing 20 adult lice on each piece of cloth in a 50-ml. beaker. In the patch tests 0.5 gram of a pyrophyllite powder containing the test material was rubbed onto a patch of knitted cotton underwear cloth 4.9 inches square (⅛ square foot), the treated

[1] Accepted for publication January 30, 1958.

[2] The authors were assisted by D. L. VanNatta in conducting tests and by P. N. Mason, S. R. Pratt, and R. B. Bonner in rearing the lice.

Table 1.—Effectiveness of different concentrations of sulfoxide in acetone, alone and in combination with rotenone at different ratios, in beaker tests against body lice.

Per Cent Sulfoxide	Per Cent Mortality Caused by Sulfoxide Alone	Per Cent Mortality[a] at Indicated Sulfoxide:Rotenone Ratio					
		40:1	20:1	10:1	5:1	2:1	1:1
5.0	95	100	100	100	100	100	100
2.5	83	98	100	100	100	100	100
1.0	62	100	95	100	98	98	90
0.5	3	95	93	93	95	92	95
.25	8	63	92	97	87	83	75
.1	7	22	35	62	72	72	65
.05	—	5	3	12	55	48	43
.025	—	3	5	3	5	2	3

[a] No mortality with rotenone alone at concentrations up to 5% in acetone or with untreated cloth.

Table 2.—Effectiveness of pyrophyllite powders containing 5% of sulfoxide, alone and in combination with different percentages of rotenone in patch tests against body lice.

Per Cent Rotenone	Per Cent Mortality on Treatments Aged Days Shown							
	Initial	7	14	22	28	42	56	92
5.0	100	100	100	100	100	100	100	100
2.5	100	100	100	100	100	100	100	98
1.0	100	100	100	100	100	100	98	98
0.5	100	100	100	100	98	100	100	95
.25	100	100	100	100	100	100	95	90
Sulfoxide alone	58	65	93	78	93	95	73	78
Pyrophyllite check	5	10	0	0	0	5	5	0

Table 3.—Residual effectiveness of 0.1% of rotenone in acetone in combination with 1% of selected compounds in beaker tests against body lice.[a]

ENT No.	Compound	Per cent Mortality Caused by Sulfoxide Alone	Per Cent Mortality on Treatments Aged for Days Shown						
			Initial	7	14	21	28	42	56
14250	Piperonyl butoxide	5	100	100	98	98	90	88	95[b]
15266	Propyl isome	0	63	—	—	—	—	—	—
18323	Benzene, 1,2-methylenedioxy-4-[2-(octylsulfonyl)propyl]-	87	100	100	97	95	93	82	87[b]
20003	Chrysanthemumic acid, *alpha*-allylpiperonyl ester	100	83	57	—	—	—	—	—
20091	Chrysanthemumic acid, *alpha*-propylpiperonyl ester	0	67	—	—	—	—	—	—
20244	Chrysanthemumic acid, 4-(3,4-methylenedioxyphenyl)-*sec*-butyl ester	87	88	83	83	80	97	77	83[b]
20272	Chrysanthemumic acid, *alpha*-ethylpiperonyl ester	0	97	87	92	80	90	70	—
20309	Chrysanthemumic acid, *alpha-tert*-butylpiperonyl ester	100	78	—	—	—	—	—	—
20313	*m*-Dioxane, 5-butyl-5-ethyl-2-(3,4-methylenedioxyphenyl)-	97	100	100	100	98	98	95	97[b]
20332	Fencholic acid, *alpha*-allylpiperonyl ester	90	98	92	88	93	95	90	92[b]
20337	Chrysanthemumic acid, *alpha*-isopropylpiperonyl ester	50	100	68	—	—	—	—	—
20425	Chrysanthemumic acid, *alpha*-(3-phenylpropyl)piperonyl ester	40	90	82	68	—	—	—	—
20426	Chrysanthemumic acid, *alpha*-cyclohexylpiperonyl ester	0	30	—	—	—	—	—	—
20616	Acetic acid, phenyl-, *alpha*-isopropylpiperonyl ester	50	100	88	80	63	—	—	—
20857	Chrysanthemumic acid, 3-(3,4-methylenedioxyphenyl)propyl ester	10	90	70[c]	—	—	—	—	—
16634	Sulfoxide (Standard)	3	100	100	100	100	100	93	93[b]
—	Untreated check	—	0	0	0	5	5	5	0
—	Rotenone alone	—	5	—	—	—	—	—	—

[a] Average of three replications of 20 lice each.
[b] Still about as effective after 90 days, when the test was terminated.
[c] After 2 days.

cloth was fastened to a plywood panel, and 20 adult lice were confined on the cloth inside a metal fruit-jar ring. Both types of tests were conducted at 80° F. and 60 to 70% relative humidity, and the lice were exposed for 24 hours. There were three replications of each test.

In sleeve tests 3 grams of pyrophyllite powder containing the test material and 3% of Celite was rubbed into knitted cotton cloth sleeves 1 square foot in area, the treated sleeves were fastened with elastic adhesive tape to the arms and legs of human subjects, and twice each week 25 adult lice were introduced into each sleeve and left for 24 hours. Sleeves were worn and tested as long as they killed 75% of the lice. Four replications were made of each test.

RESULTS.—Results of beaker tests with sulfoxide at several concentrations in combination with rotenone at different ratios are shown in table 1. Residual effectiveness in patch tests with pyrophyllite powders containing 5% of sulfoxide combined with rotenone at different concentrations are shown in table 2. In both types of tests the effectiveness of the treatment varied with the amounts of both sulfoxide and rotenone. In the beaker tests rotenone alone at concentrations up to 5% caused no mortality of lice, whereas sulfoxide alone at 5% was highly toxic.

The results of beaker tests showing the residual effectiveness of 16 selected materials combined with rotenone are presented in table 3. Piperonyl butoxide, sulfoxide, 1,2-methylenedioxy-4-[2-octylsulfonyl)-propyl]benzene, 5-butyl-5-ethyl-2-(3,4-methylenedioxyphenyl)-*m*-dioxane, the *alpha*-allylpiperonyl ester of fencholic acid, and the 4-(3,4-methylenedioxyphenyl)-*sec*-butyl ester of chrysanthemumic acid were the most effective.

The results of additional tests with various concentrations of the best of these compounds on freshly treated patches are shown in table 4. All the materials were about equally effective, but the chief effect of all except piperonyl butoxide and sulfoxide was additive, instead of synergistic, as shown by their levels of toxicity alone.

In sleeve tests against DDT-resistant lice (Korean A colony) pyrophyllite powder containing 5% of sulfoxide and 0.5% of rotenone gave 100% mortality for 17 days, and 97% to 100% for 20 to 27 days. A powder containing 2% of sulfoxide and 0.2% of pyrethrins gave 100% mortality for 10 days, 99% to 98% for 13 to 17 days, and 93% to 83% for 20 to 27 days.

It is interesting to note that in these tests rotenone alone caused little or no mortality whereas some of the piperonyl compounds alone caused high mortality at

Table 4.—Effectiveness of six piperonyl-type compounds at different concentrations in acetone combined with rotenone at a ratio of 10:1 in beaker tests.[a]

Compound	Per Cent Mortality[b] at Indicated Concentration of Compound		
	0.5	0.25	0.1
Benzene, 1,2-methylenedioxy-4-[2-(octylsulfonyl)propyl]-	97 (92)	93 (83)	43 (17)
Chrysanthemumic acid, 4-(3,4-methylenedioxyphenyl)-*sec*-butyl ester	63 (77)	55 (33)	28 (13)
m-Dioxane, 5-butyl-5-ethyl-2-(3,4-methylenedioxyphenyl)-	88 (98)	83 (58)	53 (7)
Fencholic acid, *alpha*-allylpiperonyl ester	92 (70)	80 (37)	40 (8)
Piperonyl butoxide	65 (5)	40	28
Sulfoxide	100 (3)	78	35

[a] Figures in parentheses represent mortality caused by piperonyls alone.
[b] Mortality on untreated cloth patches ranged from 0 to 5%; rotenone alone caused no mortality at these concentrations.

some concentrations. This is the reverse of the situation that exists with pyrethrins and allethrin and their synergists, at the concentrations at which they are normally used.

REFERENCES CITED

Burden, G. S., and M. M. Cole. 1956. Effectiveness of cyclethrin with various synergists against body lice. Jour. Econ. Ent. **49**(5): 643–5.

Bushland, R. C., L. C. McAlister, Jr., G. W. Eddy, H. A. Jones, and E. F. Knipling. 1944. Development of a powder treatment for the control of lice attacking man. Jour. Parasitol. **30**(6): 377–87.

Douglas, J. R., and D. P. Furman. 1949. Comparative evaluations of rotenone formulations for cattle grub control. Jour. Econ. Ent. **42**(6): 884–7.

Eddy, G. W., M. M. Cole, and A. S. Marulli. 1954a. Louse powder synergists. Soap and Chem. Spec. **30**(7): 121–3, 143, 145.

Eddy, G. W., M. M. Cole, and G. S. Burden, 1954b. Synergists with allethrin against the body louse. Jour. Econ. Ent. **47**(3): 501–6.

Moore, D. 1950. Laboratory studies of combinations of piperonyl cyclonene, piperonyl butoxide, pyrethrins, and rotenone for the control of ticks on dogs. Jour. Parasitol. **36**(4): 322–5.

Pearson, A. M. 1935. The role of pine oil in cattle fly sprays. Delaware Agric. Expt. Sta. Bull. **196.**

Pierpont, R. L. 1939. Terpene ethers in pyrethrum and rotenone fly sprays. Delaware Agric. Expt. Sta. Bull. **217.**

Roberts, I. H., J. A. Lofgren, and W. L. Berndt. 1953. Tests with insecticide formulations for the destruction of cattle grubs. Jour. Econ. Ent. **45**(6): 909–13.

Trembley, H. L. 1943. Derris used for the control of head lice and pubic lice, Jour. Econ. Ent. **36**(5): 795.

The Effect of Entry into Dieldrin-Treated Soil Upon Adults of the European Chafer[1]

H. H. SHOREY and GEORGE G. GYRISCO,[2] *Cornell University, Ithaca, N. Y.*

ABSTRACT

Adults of the European chafer (*Amphimallon majalis* (Raz.)) were introduced into cages which had been placed in untreated plots and plots treated with dieldrin granules at 2 pounds of actual toxicant per acre. The bottoms of the cages were open, and the adults were free to burrow into the soil. Many more beetles were capable of emergence from untreated than from treated soils on the days following introduction. Application of the insecticide 10 weeks or 1 year before introduction of beetles caused fewer adults to emerge than application 5 days before introduction. Males were more susceptible than were females. Although the number of eggs laid in treated soil was reduced by more than 50%, no effect of the treatment upon egg viability was detected.

Since the European chafer (*Amphimallon majalis* (Raz.), Scarabaeidae) was discovered in New York in 1940, many experiments have been conducted to find practical means by which the size of larval populations can be reduced in meadow and pasture sod (Burrage & Gyrisco 1952, 1953, Gyrisco & Evans 1954, Evans & Gyrisco 1955, Evans *et al.* 1957). The experiments have consisted of applications of various formulations and concentrations of insecticides to the soil surface, followed by soil sampling for a determination of the effect of the treatment upon densities of larval populations. Often one or more periods of European chafer adult flight and oviposition, which take place during June and July of each year, have occurred between the time of treatment of the soil and the subsequent determination of the degree of larval infestation. Possibly a portion of the decrease found in such cases has been the result of the action of insecticides upon the adults, which must leave the soil to mate and re-enter to oviposit and pass the daylight hours.

METHODS AND MATERIALS.—Two separate experiments, designated as A and B, were conducted during June and July, 1956 in a meadow located in Marion, Wayne County, New York. All tests were conducted with dieldrin[3] which was applied at the rate of 2 pounds of actual toxicant per acre with hand-operated fertilizer spreaders.

For Experiment A, 24 plots, each 10 by 20 feet, were arranged in six blocks. In order to include a variety of weather conditions and different populations of beetles in the test and thus broaden the inferences which might be made from the average results obtained, two of the blocks were devoted to each of three time intervals, which corresponded approximately to the early, middle, and latter portions of the adult flight period. Within each block 1 plot was left untreated, 1 received an application of dieldrin at 5 days, 1 at about 10 weeks, and 1 at about 1 year before exposure of the beetles. At the beginning of each time interval a screened cage measuring 2×2×2 feet was placed in the center of each of the eight plots constituting that interval. A sheet metal projection extended down 8 inches into the soil from the periphery of the open bottom of each cage to restrict the horizontal movement of European chafer adults.

The adults were collected from small trees during the evening and were divided at random and without regard to sex into lots of 350 beetles; 1 lot was placed in each cage. The introduced beetles were free to fly to and mate

[1] Presented at the Twenty-ninth Annual Meeting of the Eastern Branch of the Entomological Society of America held in New York, N. Y., November 25 and 26, 1957. Accepted for publication January 30, 1958.

[2] The authors acknowledge the assistance of W. G. Evans, now Assistant Professor, Virginia Polytechnic Institute, Blacksburg, Virginia, and E. W. Huddleston, Cornell University, Ithaca, N. Y.

[3] Granulated dieldrin, 2% active ingredient, Attapulgus AA 30–60 mesh, Shell Chemical Corporation, New York, N. Y

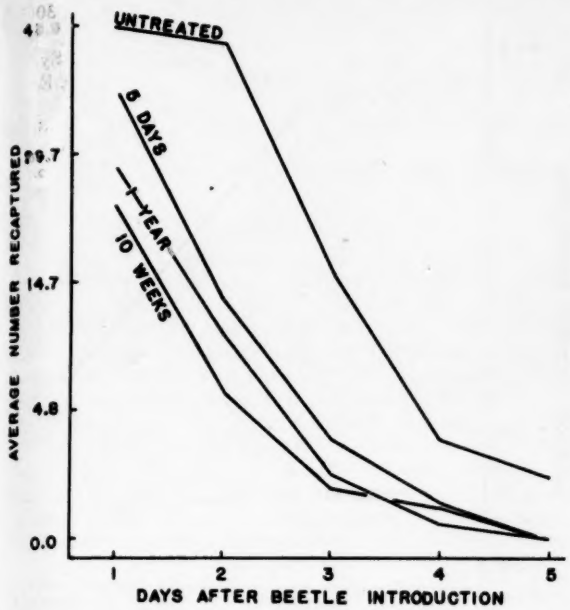


FIG. 1.—Numbers [a] of adults of the European chafer, *Amphimallon majalis* (Raz.), averaged over sexes, emerging on each of 5 consecutive days following entry into untreated soil and soils treated with dieldrin at three time intervals before entry.

[a] Vertical axes are maintained on the transformed scale: $X' = \sqrt{X+0.5}$. Averages presented are converted back to original units from the transformed data.

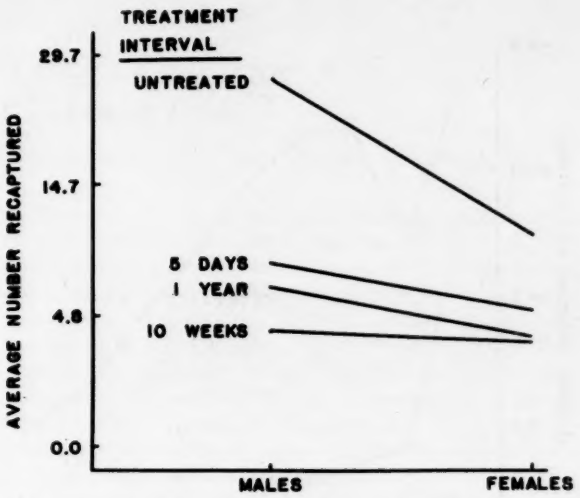


FIG. 2.—Numbers [a] of males and females of the European chafer, averaged over a 5 day period, emerging following entry into untreated soil and soils treated with dieldrin at three time intervals before entry.

upon the screen or to burrow into the soil. On the 5 evenings following introduction the adults emerged from the soil and flew at approximately the same hour as in the surrounding field. After a brief hovering flight near the sides and tops of the cages, they alighted upon and clung to the screen, and mating commenced. After emergence from the soil had almost ceased, and before many beetles had dropped from the screen, a plywood panel was inserted through a slot in the lower margin of the front of each cage to form a barrier above the soil surface. All adults within the cages above the panels were removed, counted, sex determined, and destroyed. Then the panels were withdrawn so that those beetles which had not emerged and flown could do so on some subsequent evening. The numbers of males and females recovered from the various treatments on the 5 evenings following introduction are presented graphically in figures 1 and 2.

The area for Experiment B was divided into 8 plots, each 15 by 14.5 feet, of which 4 were treated with dieldrin and 4 were left untreated. Eleven days following treatment, 200 beetles were introduced into a cage which was placed in the center of each plot. For 9 consecutive evenings all beetles which emerged were removed, counted by sex, and reintroduced into the cages. Thus a single beetle could fly more than once within a cage. The numbers of males and females recovered on each evening are illustrated in figures 3 and 4.

After the 9 days had elapsed, a square-foot hole was dug to a depth of 6 inches in the center of the area over which each cage had stood. The soil removed was sifted through screens of varying mesh sizes in order to recover all eggs, newly hatched first-instar larvae, and living adults. Most of the undamaged eggs were transferred with a camel-hair brush to squares of moistened absorbent paper covering the bottoms of small plastic specimen boxes. The boxes were maintained at laboratory conditions and examined at frequent intervals for a comparison of the percentage of hatch of eggs taken from the treated and untreated soils.

DISCUSSION OF RESULTS.—Analyses of variance for split plot within split block designs were computed for the results of Experiments A and B. Original observations were transformed for analysis ($X' = \sqrt{X+0.5}$) because the variance was seen to be proportional to the mean. All differences discussed below were found to be significant at the 5% level.

In Experiment A the insecticidal treatments caused a greater than 50% reduction in the numbers of emerging beetles. Applications of dieldrin approximately 10 weeks and 1 year before the flight were more effective, on the average, than application 5 days before the flight. It is possible that much of the insecticide had entered into only the superficial layers of the soil by the fifth day after application and that beetles in those plots were exposed to the toxicant only during the brief period when they were entering and leaving the soil.

In Experiment B natural conditions were simulated as closely as possible, with some beetles existing in an area free of insecticides and others having access to dieldrin-treated soil only. It is apparent that many of the beetles in the untreated plots flew and were counted several times, while few of those exposed to treated soil were capable of flight after the first night.

In both experiments males, although they were initially more abundant, were affected more severely by exposure to dieldrin than females.

It was not demonstrated that beetles which failed to emerge from treated soil were killed or incapacitated quickly, although such was assumed to be the case. However, almost all the adults in both treated and untreated soils were dead after 9 days, since the eight soil samples

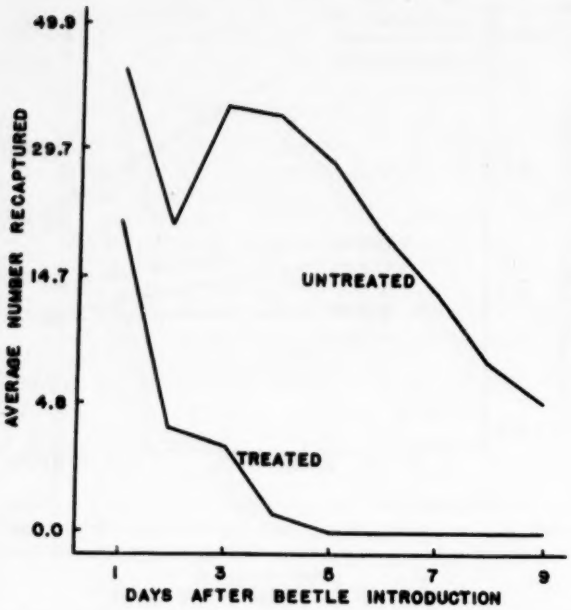


FIG. 3.—Numbers [a] of adults of the European chafer, averaged over sexes, emerging on each of 9 consecutive days following entry into untreated soil and soil treated with dieldrin.

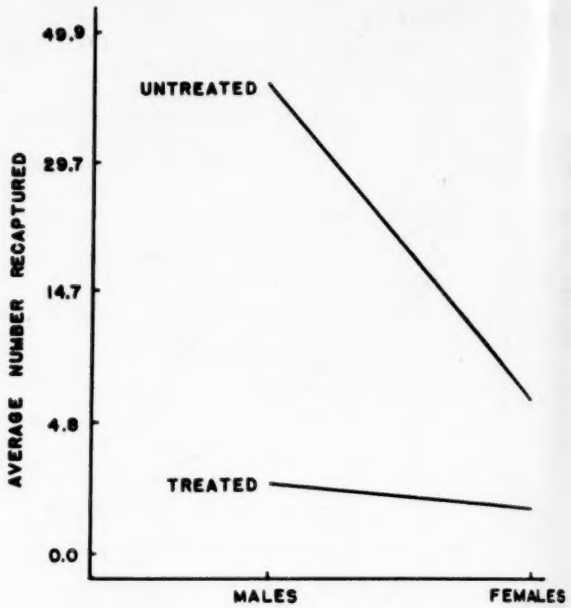


FIG. 4.—Numbers [a] of males and females of the European chafer averaged over a 9-day period, emerging following entry into untreated soil and soil treated with dieldrin.

taken following the conduct of Experiment B contained only one living female and no living males. Even if they remained alive below the soil surface for several days, females exposed to dieldrin laid fewer than half as many eggs as those not exposed. Averages of 177.8 and 80.0 eggs and first instar larvae per square foot were found in the untreated and treated plots, respectively. Therefore, at least 50% of the reduction in numbers which would have been evident during a fall sampling of larval populations was due to the effect of dieldrin upon the adults.

The percentage of hatch of the eggs which were removed from untreated and treated soils and placed among uncontaminated, favorable conditions were 89.3 and 87.8%, respectively. Because no significant reduction in the percentage of hatch was detected, no effect of dieldrin on egg viability was indicated.

REFERENCES CITED

Burrage, R. H., and George G. Gyrisco. 1952. Control of the European chafer in pasture sod. Jour. Econ. Ent. 45(2): 313–5.

Burrage, Robert H., and George G. Gyrisco. 1953. Control of European chafer damage in pasture sod—II. Jour. Econ. Ent. 46(1): 142–6.

Evans, William G., and George G. Gyrisco. 1955. Granulated insecticides for control of the European chafer in meadows. Jour. Econ. Ent. 48(4): 384–6.

Evans, W. G., H. H. Neunzig, George G. Gyrisco, and Roy G. Richmond. 1957. Aerial applications of granulated dieldrin for control of larvae of the European chafer on hilly meadows and pastures. Jour. Econ. Ent. 50(3) 273–6.

Gyrisco, George G., and W. G. Evans. 1954. Granular insecticides. Agric. Chemicals 9(1): 32–33.

NOTICE TO AUTHORS

Authors are urged to consult the Publication Policies and Manuscript Rules and Suggestions (Vol. 49(4): 575–7) before preparing manuscripts intended for publication in the JOURNAL OF ECONOMIC ENTOMOLOGY.

Control of Thrips and the Cotton Fleahopper with Sprays in Central Texas in 1957[1]

J. W. Davis, C. R. Parencia, Jr., and C. B. Cowan, Jr., *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*[2]

Abstract

The efficacy of several insecticides against thrips, *Frankliniella* sp., and the cotton fleahopper, *Psallus seriatus* (Reut.), infesting cotton was evaluated in field experiments. The addition of methyl parathion or malathion to toxaphene did not improve control of thrips over toxaphene alone. Toxaphene at 0.75 and 1.5 pounds, dieldrin at 0.1 pound, methyl parathion at 0.25 pound, Guthion at 0.13 and 0.26 pound, Sevin (1-naphthyl-N-methyl carbamate) at 0.5 pound, and Monsanto CP-7769 (diethyl (ethylthiomethylidyne)triphosphonic acid) at 0.25 pound per acre were effective against these pests. Toxaphene at 0.75 and 1.5 pounds, Guthion at 0.25 pound, and Sevin at 0.5 pound per acre gave good control of the cotton fleahopper. Thiodan at 0.5 pound per acre gave poor control of both insects

Heavy spring rains in central Texas in 1957 were responsible for a later than usual planting of the cotton crop. Because host plants were late, heavy infestations of thrips, *Frankliniella* sp., and of the cotton fleahopper, *Psallus seriatus* (Reut.), developed in this acreage. These infestations afforded an opportunity to conduct field-plot experiments with various insecticides for their control in the vicinity of Waco.

Procedure.—In experiments 1, 2, and 3 the plots were 16 rows wide and of sufficient length for an area of 0.2 acre. They were arranged in randomized blocks with four replications of treatments. Untreated plots were not included within the randomized-block experimental design, but were located in the same fields adjacent to the treated plots. In large-plot experiments 4, 5, and 6 the plots were 32 to 48 rows wide and 2 to 8 acres in size. In experiment 4 treatments were replicated twice, but in experiments 5 and 6 treatments were not replicated.

Methods of applying insecticides were the same as those reported by Davis *et al.* (1957). Insecticides for thrips control were usually applied weekly and those for cotton fleahoppers at 12 to 14-day intervals.

The procedure and method of recording thrips data were the same as those reported by Parencia & Cowan (1953) and those for the cotton fleahopper and other pests as reported by Ewing & Parencia (1943).

Experiments 1, 2, 3, and 4 were conducted for thrips control and experiments 5 and 6 for cotton fleahopper control. Cotton fleahoppers were also a factor in experiments 1 and 2. Although the bollworm, *Heliothis zea* (Boddie), occurred in all plots, it was considered of importance only in experiments 3, 4, and 6. Infestations of the boll weevil, *Anthonomus grandis* Boh., were light and did not appreciably affect results in any of the experiments.

The insecticides used in these experiments are listed in tables 1 and 2. The emulsifiable concentrates were either commercial products or experimental materials obtained from various manufacturers. Noticeable phytotoxicity occurred from spray applications of Sevin in all experiments in which it was used.

Results.—In experiment 1 both dosages of toxaphene, methyl parathion alone, and toxaphene plus methyl parathion or malathion gave significantly better thrips control than malathion alone. The addition of the phosphorus compounds to toxaphene did not improve control over toxaphene alone. There was no difference in cotton fleahopper control among insecticides. Yields were severely reduced by dry weather during the main fruiting period. The 1.5-pound dosage of toxaphene and the toxaphene plus malathion mixture gave significantly higher yields than methyl parathion or malathion.

In experiment 2 each of the other insecticides gave significantly better thrips control than Thiodan. Guthion at the 0.26-pound dosage was better than dieldrin. Sevin (1-naphthyl-N-methyl carbamate) and the lower dosage of Guthion were better than Monsanto CP-7769 (diethyl (ethylthiomethylidyne)triphosphonic acid). Dieldrin, both Guthion dosages, and Sevin gave better cotton fleahopper control than Thiodan. Dieldrin and the higher dosage of Guthion gave significantly higher yields than the other insecticides.

[1] Accepted for publication January 31, 1958. Presented at the meeting of the Southwestern Branch, Entomological Society of America, held at Houston Texas, February 10–11, 1958.

[2] In cooperation with the Texas Agricultural Experiment Station.

Table 1.—Effectiveness of several insecticides against thrips on cotton.

Insecticide (Pounds per Gallon)	Active Ingredient (Pounds per Acre)	Average Number of Thrips per Plant	
		Before Treatment	After Treatment
Experiment 1 (Spray applications on June 7, 14, and 25)			
Toxaphene 6	0.75	4.9	1.2
	1.5	3.1	1.3
Toxaphene 6+methyl parathion 2	.75+0.25	4.7	0.8
Methyl parathion 2	.25	4.8	1.4
Toxaphene 6+malathion 5	.75+ .31	4.3	1.0
Malathion 5	.31	4.5	5.0
L.S.D. at 5% level	—	—	1.2
Untreated outside check	—	5.1	8.8
Experiment 2 (Spray applications on June 7, 14, and 25)			
Dieldrin 1.5	0.1	3.1	1.3
Guthion 1.5	.13	3.8	0.7
	.26	4.1	0.4
Thiodan 2	.42	3.2	4.0
Sevin 1	.5	3.0	0.6
CP-7769, 2	.125	5.9	1.6
L.S.D. at 5% level	—	—	0.8
Untreated outside check	—	3.7	8.0
Experiment 3 (Spray applications June 27, July 4 and 11)			
Dieldrin 1.5	0.1	5.6	3.0
Sevin 1	.25	7.6	4.4
	.5	7.2	2.4
Thiodan 2	.5	8.2	6.2
CP-7769, 2	.125	8.0	3.9
	.25	7.6	1.8
L.S.D. at 5% level	—	—	0.7
Untreated outside check	—	9.9	13.8
Experiment 4 (Spray applications on June 27 and July 4)			
Toxaphene 6	0.9	11.2	2.8
Guthion 1.5	.13	7.6	1.4
	.26	9.7	0.6
Untreated check	—	8.2	8.2

Table 2.—Cotton fleahopper, boll weevil, bollworm infestation and yield records in thrips and cotton fleahopper-control experiments.

Insecticide (Pounds per Gallon)	Active Ingredient (Pounds per Acre)	Flea-Hoppers per 100 Terminals	Per Cent of—			Pounds of Seed Cotton per Acre	
			Weevil-Punctured Squares	Bollworm-Injured Squares	Bollworm-Injured Bolls	Total	Gain Over Check
Experiment 1 (Spray applications June 7, 14, and 25)							
Toxaphene 6	0.75	16.3	8.5	4.4	3.6	390	26
	1.5	12.8	7.1	4.0	3.3	426	62
Toxaphene 6+methyl parathion 2	.75+0.25	14.4	7.5	3.9	2.7	421	57
Methyl parathion 2	.25	16.8	6.7	3.2	2.4	359	−5
Toxaphene 6+malathion 5	.75+ .31	13.3	7.1	3.8	4.0	432	68
Malathion 5	.31	16.4	7.8	3.2	2.8	380	16
L.S.D. at 5% level	—	—	—	—	—	45	—
Untreated outside check	—	31.5	16.8	5.0	4.5	364	—
Experiment 2 (Spray applications June 7, 14, and 25)							
Dieldrin 1.5	0.1	12.8	6.2	2.9	1.2	522	208
Guthion 1.5	.13	15.1	6.6	1.8	1.0	454	140
	.26	12.8	5.9	2.2	1.5	524	210
Thiodan 2	.42	20.8	7.1	2.7	1.7	463	149
Sevin 1	.5	15.1	5.3	2.5	1.6	457	143
CP-7769, 2	.125	17.0	5.5	2.2	0.7	456	142
L.S.D. at 5% level	—	4.3	—	—	—	55	—
Untreated outside check	—	31.3	12.3	4.6	1.8	314	—
Experiment 5 (Spray applications June 11 and 25)							
Guthion 1.5	0.2	18.8	7.1	2.3	2.4	628	315
Toxaphene 6	1.1	17.4	7.4	2.3	4.4	574	261
Untreated check	—	36.8	21.1	2.2	2.8	313	—
Experiment 6[a] (Spray applications June 20 and July 2)							
Toxaphene 6	1.5	14.7	5.5	5.5	4.3	617	561
	0.75	20.6	5.5	6.3	5.8	551	495
Sevin 1	.5	14.8	5.4	6.3	6.1	543	487
	.25	24.5	5.8	7.5	5.5	502	446
Thiodan 2	.5	35.8	5.6	6.5	6.0	501	445
	.25	35.1	5.5	7.6	8.5	486	430
Guthion 1.5	.25	17.0	4.7	7.1	6.1	607	551
CP-7769, 2	.25	24.3	4.2	6.7	8.1	520	464
Untreated check	—	45.9	9.0	15.0	25.3	56	—

[a] Fleahopper records made before treatment for bollworm control.

In experiment 3 each of the other insecticides gave significantly better thrips control than Thiodan. Dieldrin, Sevin at 0.5 pound, and CP-7769 at 0.25 pound were better than Sevin at 0.25 pound and CP-7769 at 0.125 pound per acre. CP-7769 at the 0.25-pound dosage was better than dieldrin. A heavy bollworm infestation developed when plants began to fruit. All plots were treated eight times between July 18 and August 20 with the toxaphene-DDT spray mixture for bollworm control. Because this treatment was begun when the plants were so young, yields could not be expected to reflect differences in thrips control; therefore, they are not included in table 2.

In experiment 4 both Guthion dosages gave better thrips control than toxaphene. A bollworm infestation developed when plants began to fruit, and the experiment was converted into one for bollworm control.

In experiment 5 there was no appreciable difference in cotton fleahopper control or yield obtained with Guthion or toxaphene. Both gave good increases in yield over check.

In experiment 6 both dosages of toxaphene, Sevin at 0.5 pound, and Guthion at 0.25 pound per acre gave good cotton fleahopper control. Fair control was obtained with 0.25-pound dosages of Sevin and CP-7769. Thiodan at 0.25 and 0.5 pound gave poor control. A heavy bollworm infestation developed in the experiment, and all plots except the check received six applications of 0.4 pound of dieldrin plus 1.5 pound of DDT between July 9 and August 1 and two applications of BHC at 0.45 pound of gamma isomer plus 1.25 pound of DDT per acre on August 8 and 12. This control favored the weaker cotton fleahopper treatments in yield, because it permitted plants to replace most of the fruit lost to the cotton fleahopper. However, yields were somewhat higher in the better treatments. Yield in the check was low, as bollworms removed most of the fruit which was missed by cotton fleahoppers.

References Cited

Davis, J. W., C. B. Cowan, Jr., and C. R. Parencia, Jr 1957. Thrips control on cotton with phosphorus insecticides. Jour. Econ. Ent. **50**(5): 676–8.

Parencia, C. R., Jr., and C. B. Cowan, Jr. 1953. Field tests with sprays for the control of thrips and the cotton fleahopper. Jour. Econ. Ent. **46**(4): 633–8.

Ewing, K. P., and C. R. Parencia, Jr. 1943. Dosages of insecticides to control the boll weevil and the bollworm. Jour. Econ. Ent. **36**(4): 607–10.

Estimation of Red Clover Yield Losses Caused by the Clover Root Borer[1]

K. P. Pruess and C. R. Weaver[2]

Abstract

Reductions in second cutting red clover hay yields caused by the clover root borer, *Hylastinus obscurus* (Marsham), were estimated by multiple regression analyses. Variables included were number of borers, crown diameter of the root, the square of crown diameter, and foliage weight. Both foliage weight and number of borers per root were found to increase as crown diameter increased, the relationships being curvilinear. Regression of foliage weight on borers, independent of crown diameter, showed an increase in number of borers to result in a decrease in foliage weight. Calculations indicated that an average of 1.5 borers per root resulted in a 5.5% reduction in yield in Ohio in 1956.

The clover root borer, *Hylastinus obscurus* (Marsham), is one of the most serious insect pests of red clover. Various workers (Weaver & Haynes 1955, Woodside & Turner 1956, Weaver *et al.* 1957) have reported yield increases resulting from clover root borer control. However chemicals used for clover root borer control also reduce populations of many other insects feeding on red clover. For this reason, yield increases resulting from chemical treatments may not be entirely attributable to control of a specific insect species.

The best method of estimating root borer damage appeared to be to determine the effect of root borers on individual plants in untreated fields, assuming that the presence of borers is independent of other factors reducing red clover yields.

Methods.—As part of a sampling study, 320 red clover plants were systematically dug from each of three medium red clover fields. Samples were taken between Aug. 7 and Aug. 22, 1956 at the time the second cutting of red clover hay was being made. First cutting hay yields are seldom affected by the clover root borer and a third cutting of red clover hay is rarely obtained in Ohio. Therefore yield reductions caused by the clover root borer are manifest almost entirely in the second cutting.

Green weight of the foliage of each plant was recorded to the nearest gram, the diameter of each root was measured at the crown to the nearest millimeter, and each root dissected to determine the number of borers present. Fields 1 and 2 were located on the Apple Creek State Hospital Farm in Wayne County while Field 3 was on the Orlo Blackstone farm 4 miles southeast of Dalton in Stark County, Ohio. Most root borers were full grown larvae, pupae, or newly emerged adults at the time samples were taken and feeding injury had largely ceased. However a few eggs were found in roots on the Blackstone farm and some small larvae were still to be found in all fields. Relationships among number of borers, crown diameter, and foliage weight were determined by correlation analyses. Since there were large differences among the fields in borer populations and plant size, field to field variation was removed when combining data from the three fields.

Results.—Table 1 summarizes these correlations. Simple correlation between number of borers and foliage weight showed weight to increase slightly as borers increased. However number of borers and foliage weight were both related to root size, both increasing as the crown diameter increased. These relationships were both curvilinear, second degree curves significantly increasing the correlation coefficients. Third degree curves failed to improve either relationship.

To test more accurately the relationship between foliage weight and number of root borers, a multiple regression analysis was used to remove the root size variable. The square of crown diameter was included as a fourth variable to compensate for the curvilinear relationships between crown diameter and foliage weight and between crown diameter and number of borers. In analyses of data from individual fields, inclusion of this fourth variable significantly increased the multiple correlation coefficients. However when data from the three fields were combined in a single analysis, inclusion of this variable increased the multiple correlation coefficient only from 0.5693 to 0.5699 which was not significant. Nevertheless, it seems appropriate to retain this variable because of the curvilinear relationships previously noted.

A brief summary of data from the individual fields and estimates of yield reduction obtained by multiple regression methods are presented in table 2. Because of the variable nature of the data used in these analyses, the partial regression coefficient for weight on borers was significant only for Field 2. However this coefficient was also negative for the other two fields, indicating a tendency for yields to decrease as the number of borers increased.

In an effort to obtain a better estimate of losses by the clover root borer, and to obtain a formula applicable to the general situation, data from these three fields were incorporated into a single analysis. This analysis resulted in a highly significant partial regression coefficient for weight on borers, giving an estimate of 2.0140 ± 1.2296

[1] Accepted for publication February 11, 1958.

[2] Research Assistant and Associate Entomologist, respectively, Ohio Agricultural Experiment Station, Wooster. The senior author is now Assistant Entomologist, University of Nebraska, North Platte Experiment Station, North Platte.

Table 1.—Correlation coefficients.

Variables Correlated	Correlation Coefficients	
	Linear	Curvilinear
Crown diameter and borers	0.2735**	0.2821[a]
Crown diameter and weight	0.5639**	0.5693[a]
Borers and weight	0.0637*	—

* Significant at the .05 level.
** Significant at the .01 level.
[a] Significantly greater than linear correlation coefficient.

Table 2.—Yield reductions caused by clover root borer.

Field	Borers per Root	Mean Crown Diameter (Mm.)	Mean Foliage Weight (Gm.)	Estimated Mean Weight in Absence of Root Borers	Per Cent Reduction in Yield
1	0.84	14.33	110.37	112.93	2.3
2	1.49	10.78	33.50	36.01	7.0
3	1.26	9.56	28.20	28.97	2.7

grams reduction in foliage weight per plant per borer.

The multiple regression equation resulting from this analysis gave $Y = -7.0030 + 4.1254X_1 + 0.1285X_2 - 2.0140X_3$ where Y is foliage weight in grams, X_1 is crown diameter in mm., X_2 is the square of crown diameter, and X_3 is number of root borers. Substituting different values for crown diameter and borers into this equation, it is possible to estimate per plant yields for any number of borers and to compare this value with the estimated yield for uninfested plants of the same size. Figure 1 summarizes the estimates of yield reduction caused by varying numbers of borers in different sized plants.

Samples taken from 11 fields in Ohio in 1956 showed an average of 1.5 borers per root while mean crown diameter, based on samples from six fields, was 11.2 mm. Using either the regression equation or our graph, it is estimated that a yield reduction of about 5.5% occurred due to the clover root borer. Clover root borer populations were much lower than normal in Ohio in 1956 while weather conditions were unusually favorable for red clover growth. Further work would be desirable to determine losses under varying soil moisture conditions, cutting times, and at different stages in borer development.

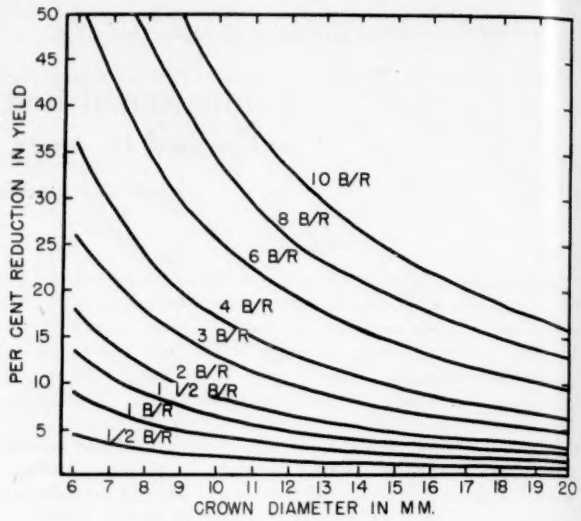


FIG. 1.—Per cent reduction in yield of second cutting red clover caused by different numbers of clover root borers per root (B/R) in different sized plants.

REFERENCES CITED

Weaver, C. R., and J. L. Haynes. 1955. Band placement of insecticides for clover root borer control. Jour. Econ. Ent. **48**: 190–1.

Weaver, C. R., K. P. Pruess and J. L. Parsons. 1957. Further tests of band placement of insecticides for clover root borer control. Jour. Econ. Ent. **50**: 255–6.

Woodside, A. M., and E. C. Turner, Jr. 1956. Control of the clover root borer in Virginia. Jour. Econ. Ent. **49**: 640–3.

Control of Insects Affecting Birdsfoot Trefoil Seed Production in Vermont[1]

GEORGE B. MACCOLLOM,[2] *Vermont Agricultural Experiment Station, Burlington*

ABSTRACT

The results of the two years' studies on the control of insects affecting birdsfoot trefoil (*Lotus corniculatus* (L.)) seed production show that early season applications of insecticides with good residual properties result in significant increases in seed yields. The data indicate that control of the meadow spittlebug (*Philaenus leucophthalmus* (L.)) and several species of Miridae is necessary during the critical period of bud development and flowering. The flowering of treated plots was visibly more dense, uniform, and of shorter duration than on the check plots. This observation was confirmed by counts of umbels per stem and pods per umbel, and was considered a good criterion for measuring plant protection offered by the insecticides. The honeybee population on the test plots was considered adequate for pollination.

The importance of birdsfoot trefoil, *Lotus corniculatus* (L.), as a legume forage and as a cash seed crop has increased rapidly since 1952. Observations during the past several years indicated that the activity of injurious insects and pollinators was one of the biggest single factors limiting seed production in Vermont. Of the many insect species found associated with birdsfoot trefoil, several were considered as present in sufficient numbers to cause economic losses. High nymphal populations of the meadow spittlebug, *Philaenus leucophthalmus* (L.), are found consistently each spring on birdsfoot trefoil. Following seed harvest, from midJuly to August 1, the trefoil makes a rapid recovery. Prior to frosts in early to mid-September, the regrowth is succulent and generally in partial flowering. This succulent growth, according to

[1] Vermont Agricultural Experiment Station Journal Series No. 79. Accepted for publication February 12, 1958.

[2] The author gratefully acknowledges the assistance of P. W. Oman, Director, and others of the Insect Identification and Parasite Introduction Laboratories, U. S. Department of Agriculture, Beltsville, Md., in identifying insect specimens.

Table 1.—Effect of applications of insecticides on Mansfield birdsfoot trefoil grown for seed.[a]

Insecticide	Pounds Active Ingredient/A.	Pounds Clean Seed/A.		Per Cent Control Spittlebug Nymphs 1956	Mean Number			
					Umbels/Stem		Pods/Umbel	
		1956	1957		1956	1957	1956	1957
Endrin	0.20	334.0	333.0	100	3.4	3.7	3.4	2.9
Toxaphene	2.25	336.7	279.5	95	3.2	3.1	3.5	3.1
DDT	1.00	336.3	291.9	73	2.6	3.2	3.3	3.1
DDT and Dieldrin	1.00 + 0.25	314.5	304.2	100	2.8	2.9	3.6	3.1
Check	—	165.0	218.1	—[b]	2.26	2.70	2.55	3.17
L.S.D. at 5%	—	31.76	27.10	—	.59	n.s.	.39	n.s.
at 1%	—	44.54	n.s.	—	.82	n.s.	.55	n.s.

[a] Application dates May 29, 1956; May 8, 1957. Harvest dates July 27, 1956; July 17, 1957.
[b] Check averaged 0.88 nymph per stem on June 19, 1956.

Weaver & King (1954), makes optimum conditions for oviposition by meadow spittlebug adults in the fall, thus giving rise to the large nymphal populations in the spring.

The following species of Miridae were also collected from large populations: the tarnished plant bug, *Lygus lineolaris* (P.de B.); the alfalfa plant bug, *Adelphocoris lineolatus* (Goeze); the rapid plant bug, *Adelphocoris rapidus* (Say); and *Plagiognathus* spp. The abundance of the several species of Miridae varied widely from field to field. Observations and sweeping with an insect net indicated that, on smaller acreages, populations were lower and injury was not as acute. Hixson (1955) reported that this was common on Nebraska alfalfa seed fields.

It is well known that the species listed and others cause serious depredations to alfalfa and other legume seed production. Hughes (1943) is only one of many workers who have reported on this. Although a complete evaluation of the individual roles played by injurious species has not been concluded, it was felt that, collectively, the insects mentioned were causing severe economic losses to seed producers. Neunzig & Gyrisco (1955) reported that injurious species representing all major orders except Diptera could be found on birdsfoot trefoil. Little work, however, has been done on the control of insects affecting seed production. In view of the need for adequate information on this problem, studies are in progress at the Vermont Agricultural Experiment Station to obtain data on the life histories of the insects involved, measurement of their damage, and on their control. This paper reports progress in measurement of damage and in control by insecticides.

Procedure.—Field tests were conducted in 1956 and 1957 on a 35-acre field of Mansfield birdsfoot trefoil planted in May, 1954, which has been in production of registered seed since 1955. Insecticides used, rate per acre, and application dates are given in table 1. All sprays were prepared from emulsifiable concentrates and applied with a 20-foot boom and a 44K Bean pumping unit mounted on a 4-wheel drive, ½-ton pickup truck. All applications were made at the rate of 20 gallons per acre at 4 m.p.h. with 30 pounds pressure.

The experimental design was a 5×5 Latin square with cells measuring 20 feet wide and 100 feet long. A 5-foot alley running lengthwise in the center of each cell was reserved for making insect counts and sweepings. Any damaged growth was thus confined to that area and not included in stem or harvest samples.

Periodic sweepings of each cell were taken for insect counts, as well as weekly counts of the spittlebug nymphs per stem. Assessment of spittlebug nymphal population was determined by placing a 5-foot stick, with markings every 6 inches, in the plot. The stem nearest each mark was examined and recorded. The stick was placed at random in the alley of each cell, two times for each count.

Injury from the insect species listed is manifested in flagged terminals, bud blast, blossom drop, and withered, shrunken pods. From this known injury, it was decided that stem counts of umbels would be a good criterion for measuring this damage. Fifty stems were cut at random from each cell during bloom for counts of umbels per stem, and during post-bloom for counts of pods per umbel.

Seed yields were obtained by cutting five samples from each cell. These samples were not taken at random, but rather from areas at random within the cell judged to be of uniform vegetative growth. Although this may not be considered the best practice from a biometric standpoint, sampling in this manner was necessary in order that data would not include lodged, trampled or thin stand portions of the cells. This sampling method is discussed in part by Snedecor (1946). The yield samples consisted of all plants within a circular iron hoop covering 9.603 square feet. The samples could then be weighed in grams and multiplied by 10 to give a measure of pounds per acre. The use of the iron hoop was described to the author by members of the Vermont Agricultural Experiment Station Agronomy Department. The plants within the hoop were cut by hedge trimmers, bagged, then brought to Burlington and placed in drying cabinets at 110° F. for 36 hours. The seed was removed from the samples by threshing and screening, then cleaned by the use of a small "up-hill" cleaner.

Bee density was determined by estimating the bee population on square-yard-areas between the hours of 10 a.m. and 3 p.m. during bloom. In 1957, stakes were placed at varying distances from the bee yard, and 25 counts were made in the vicinity of the stakes every 4 days during bloom.

Results and Discussion.—Two years' investigations on control of insects affecting birdsfoot trefoil seed production show that seed yields are substantially increased, as shown in table 1. The yields as shown in table 1 are not indicative of commercial production; pod shattering and combining result in 20 to 25% loss of seed present at harvest. The data from 1956 are generally more informative

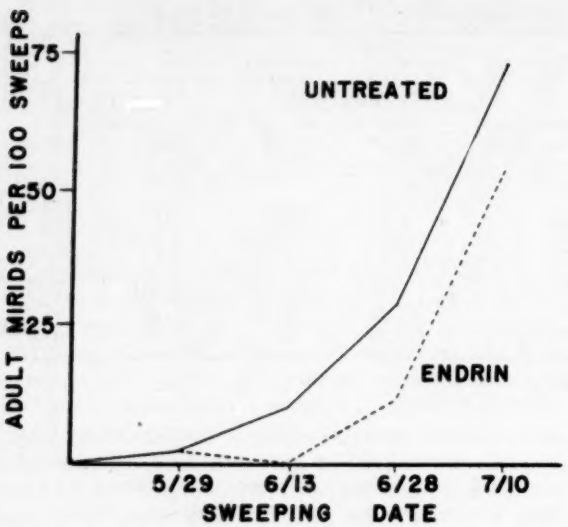


FIG. 1.—Effect of one endrin spray application on several spp. of the Miridae population. 1956.

than the data from 1957 due to climatic conditions prevailing in the spring of 1957. In 1956, the application date of May 29, two weeks prior to the start of first flowering, found the spittlebug nymphal population at its peak. The insecticides used during these studies were selected on the basis of good residual properties. The control of the meadow spittlebug, as shown in table 1, and of species of Miridae as shown in figure 1, was sufficient to protect the trefoil through bud development, bloom and pod growth, thus resulting in significant increases of umbels per stem and of pods per umbel. The flowering of the treated plots was visibly more dense, uniform, and of shorter duration than in the untreated check plots. In 1957, the earlier application date of May 8 was selected, since spittlebug hatching had been observed for 4 days, and the maximum temperature had reached 70° F. for 2 consecutive days. These optimum conditions for meadow spittlebug hatching and plant growth, however, were followed by 15 days of cool weather during which 1 day only, May 15, recorded a maximum temperature of 70° F. or above. In addition, 8 hours of freezing occurred on the morning of May 17, with a low of 24° F. Not until May 24 were there

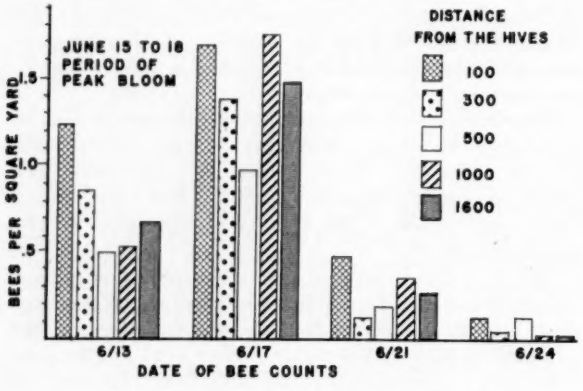


FIG. 2.—Honeybee density on birdsfoot trefoil during bloom. 1957.

Table 2.—Composition of Miridae population by species on July 10, 1956.

SPECIES	PER CENT
Lygus lineolaris	32
Adelphocoris lineolatus	25
Adelphocoris rapidus	7
Plagiognathus spp.	23
Other	13

again consecutive days of 70° F. weather. According to Weaver & King (1954), when the spring weather is variable and short cold periods are interspersed with warm periods, the hatching is prolonged over a long period. In addition, the growth of the trefoil was not sufficient to create an optimum humid microclimate for maximum nymphal survival. As a result, only partial control was derived from the May 8 applications. This is shown in table 1 by the fact that there are no significant differences in the mean number of umbels per stem or of the mean number of pods per umbel for 1957.

Although there is no analysis by percentage of the damage attributable to the meadow spittlebug or to the several species of Miridae, the data indicate that control of Miridae spp. (see table 2) is also necessary during bud development and bloom. These studies are being continued to determine satisfactory timing of applications, effects on the individual species, and benefits to be derived from control during the post-bloom period.

Throughout the course of these studies in both years, the honeybee, *Apis mellifera* (L.), population was judged satisfactory for adequate pollination. Morse (1956) states that honeybee populations of one bee per square yard are sufficient to pollinate all the birdsfoot trefoil in a given area. On the field where the tests were conducted, 41 hives of honeybees were maintained in an enclosed bee yard on the southwest corner. In 1956, square-yard counts during bloom consistently recorded more than one bee per square yard on and around the test plots. In 1957, more detailed counts were taken to determine the density of the honeybee population at varying distances from the bee yard. As shown in figure 2, one or more bees were present as far as 1600 feet from the hives during peak bloom. The farthest that honeybees had to travel for pollination was 1600 feet. These data indicate that pollination was not a factor in the results obtained from the spray tests described here.

REFERENCES CITED

Hixson, E. 1955. Insects that affect alfalfa seed production in Nebraska. Neb. Agric. Expt. Sta. Bull. **433.**

Hughes, J. H. 1943. The alfalfa plant bug. Minnesota Agric. Expt. Sta. Tech. Bull. **161.**

Morse, R. A. 1956. The pollination of birdsfoot trefoil in New York State. Abst. 10th Internatl. Congr. Ent. Sect. 15, Apiculture, #112.

Neunzig, H. H., and G. G. Gyrisco. 1955. Some insects injurious to birdsfoot trefoil in New York. Jour. Econ. Ent. **48**(4): 447–50.

Snedecor, G. W. 1946. Design and analysis of samplings. Statistical Methods, Chap. 17, 4th Ed., Iowa State College Press.

Weaver, C. R., and D. R. King. 1954. Meadow spittlebug. Ohio Agric. Expt. Sta. Res. Bull. **741.**

Effects of Systemic Insecticide Seed Treatments on Two Varieties of Cotton[1]

R. C. Dobson, *New Mexico Agricultural Experiment Station, State College*

Abstract

Thimet and Bayer 19639 were tested to determine their effects on Pima S-1 and Acala 1517C cotton. Both materials reduced seedling stands of Pima S-1 while the Thimet retarded blossoming and reduced growth during the early part of the season. The Acala 1517C reacted in a somewhat different manner to the treatments but neither variety showed a decrease nor increase in yield of cotton at the end of the season.

Systemic insecticides as seed treatments for cotton have been used for several years on an experimental basis and for the last 2 years on a limited commercial basis. Reports on possible effects on stands and yields of these seed treatments have been made from several areas (Hacskaylo 1957, Hacskaylo & Clark 1957, Parencia *et al.* 1957, Reynolds *et al.* 1957).

Since conflicting data are reported from these areas, it was deemed advisable to conduct tests on locally grown varieties under New Mexico conditions. Greenhouse tests were run with the two major varieties grown, Acala 1517C and Pima S-1, to determine any possible effects the systemics, Thimet and Bayer 19639, might have on emergence or initial growth. No significant differences were obtained in these tests which were run during December 1956, and so field plots were set out the following spring.

Methods and Procedures.—*Experiment 1.*—Pima S-1 cotton seed, acid-delinted and hot water-treated, was treated with two different systemic insecticides at the rate of 1 pound of actual toxicant per 25 pounds of seed. Thimet, as a 44% actual toxicant in carbon, was applied as a slurry. Bayer 19639 was applied in two formulations —one a 50% actual in carbon and the other a 50% emulsion. The carbon material was applied as a slurry while the emulsion was applied as a spray. All treatments were made in a hand-crank, offset-drum type mixer. All seed was treated, airdried, and bagged immediately after treatment.

Seed was planted within 2 hours after treatment in an attempt to avoid possible phytotoxic affects from storage. All planting was done with a John Deere 4-row planter at 22 pounds of seed per acre; no thinning was done. Each plot consisted of two rows of cotton planted the full length of the field and with four replications across the field. The plots were located in the middle of a 15-acre field of cotton so there was enough buffer area on each side of the plots to keep border effect at a minimum. The planting was done on April 16, 1957.

Experiment 2.—This experiment was similar to Experiment 1 in the materials used, plot design, planting methods, and equipment used. Counts and readings were also made in the same manner. The cotton seed used in this experiment was Acala 1517C, a variety developed by the Agronomy Department of the New Mexico School of Agriculture and Mechanic Arts. The seed was acid delinted. The cotton in these plots was planted on April 16, 1957. All counts were made in the same manner as those of Experiment 1.

Results.—*Experiment 1.*—Stand counts of the Pima S-1 plots were made five times during the season. The first was taken on April 29, 13 days after planting. Additional counts were made on May 15, June 5, July 11, and July 17. Analysis of the data obtained showed that there were significantly fewer plants in the treated plots than in the untreated check until the July 17 counts when no significant differences were apparent (table 1). These counts were not taken from the same places in the plots each time so counts cannot be compared from one date to the next. Figures from each treatment of each date should only be compared with the other treatment figures of that same date.

Data on numbers of blossoms per 10 plants taken on July 17, showed that Thimet-treated seed had produced significantly fewer blooms at this time than the untreated checks (table 3). The Bayer 19639-treated plants showed no retarding of the bloom.

Height counts were made twice, once on June 7 and again on June 26. No significant differences were apparent at either time between the treatments and the checks (table 2).

Early emergence observations indicated that plants from the emulsion Bayer 19639-treated plots were smaller and the appearance of the first true leaves was hastened. This obvious stunting was no longer apparent by the time the height data were taken.

Since an increase in damping-off had been suggested by other reports, data on this problem were taken. Analysis

[1] Journal Series No. 121, Agricultural Experiment Station, New Mexico College of Agriculture and Mechanic Arts, State College. Accepted for publication February 13, 1958.

Table 1.—Average number of plants per 10 feet of row in Experiment 1.

Treatment	Dates				
	4/29	5/15	6/5	7/11	7/17
Bayer 19639 50% Carbon	23.1*	32.6*	34.6*	38.1	41.3
Bayer 19639 50% Emulsion	19.5*	31.3*	31.2*	32.9*	40.7
Thimet 44D	25.7	31.4*	31.2*	33.6*	40.4
Untreated	29.9	39.2	40.6	43.1	45.3
L.S.D. at 5% level	5.51[a]	6.96[b]	5.18[a]	6.86[c]	11.50[c]
at 1% level	10.12	12.78	9.51	12.49	21.10

[a] Based on 10 samples per plot.
[b] Based on eight samples per pot.
[c] Based on four samples per plot.
* Significant at the 5% level.

Table 2.—Average height in inches of 10 plants per plot in Experiment 1.

Treatment	Dates 6/7	6/26
Bayer 19639 50% Carbon	31.8	75.0
Bayer 19639 50% Emulsion	31.2	74.3
Thimet 44D	30.1	73.0
Untreated	31.4	80.3
L.S.D. at 5% level[a]	4.28	7.60
at 1% level[a]	7.85	14.00

[a] Based on five samples per plot.

of these data, taken on May 15, showed neither an increase nor a decrease in the incidence of damping-off (table 3).

The first picking was on November 6. Analysis of these data show that the treatments applied at seeding time did not increase or decrease the production of cotton in this test (table 3).

Experiment 2.—Stand counts of the Acala 1517C cotton were taken four times during the season—May 2, May 16, June 7, and July 17. Analysis of the data obtained showed the Bayer 19639-emulsion treatment reduced the stand from the first through the third reading. The Bayer 19639 carbon treatment reduced the stand significantly below the check in the second and third counts while a reduction in stand was apparent in the Thimet-treated plots in the third count. By the fourth count, on July 17, there were no significant differences apparent between any of the treatments and the check (table 4). This was thought due to natural crowding out of weaker plants.

Height measurements were made June 7, June 26, and July 25. These data showed that a definite stunting occurred in the Bayer 19639-emulsion treated plots through the second counts. This stunting was significant at the 5% level when compared with the check plots. By the third count on July 25, these plots had evidently recovered and no significant differences between the treatments and the checks (table 5) were evident.

Bloom counts taken on July 17 show significantly fewer blossoms present in the Bayer 19639-emulsion and Thimet-treated plots when compared with the other plots. Counts taken on July 25 show no difference in numbers of blossoms regardless of the treatment (table 6).

Yields of these plots were taken at the end of the season. Data taken on September 25 showed that the first-picking yield of the Bayer 19639-emulsion-treated plot was significantly lower than the yield of the check (table 6). There was no difference between the other treatments and the check at this picking.

The second picking was on November 12, and at this time the Thimet-treated plots produced significantly more cotton than the other three sets of plots. No explanation is offered for this. When the two pickings were combined and the total weights analyzed, no significant

Table 4.—Average number of plants per 10 feet of row in Experiment 2.

Treatment	Dates 5/2	5/16	6/7	7/17
Bayer 19639 50% Carbon	32.3	44.5*	33.4*	44.8
Bayer 19639 50% Emulsion	27.5*	48.3*	36.5	45.4
Thimet 44D	32.6	47.0*	40.2	42.3
Untreated	38.9	54.4	45.7	44.4
L.S.D. at 5% level	8.36[a]	6.05[b]	9.80[c]	22.41[d]
at 1% level	15.35	11.09	17.99	41.13

[a] Based on 10 samples per plot.
[b] Based on eight samples per plot.
[c] Based on six samples per plot.
[d] Based on four samples per plot.
* Significant at the 5% level.

Table 3.—Number of blooms per 10 plants and incidence of damping-off and yield per 10 feet of row of Pima S-1 cotton in Experiment 1.

Treatment	Damping-Off 5/15	Bloom 7/17	Yield (lbs) 11/6
Bayer 19639 50% Carbon	3.0	3.6	0.7
Bayer 19639 50% Emulsion	4.2	3.6	0.7
Thimet 44D	4.0	2.5*	0.7
Untreated	3.4	5.2	0.8
L.S.D. at 5% level	2.18[a]	2.10[b]	0.19[b]
at 1% level	4.01	3.86	0.34

[a] Based on eight samples per plot.
[b] Based on four samples per plot.
* Significant at the 5% level.

Table 5.—Average height in inches of 10 plants per plot in Experiment 2.

Treatment	Dates 6/7	6/26	7/25
Bayer 19639 50% Carbon	33.7	65.9	222.8
Bayer 19639 50% Emulsion	29.5*	58.5*	223.5
Thimet 44D	32.1	66.4	223.8
Untreated	33.7	68.8	219.8
L.S.D. at 5% level	4.15[a]	7.12[a]	25.99[b]
at 1% level	7.61	13.02	47.70

[a] Based on five samples per plot.
[b] Based on three samples per pot.
* Significant at the 5% level.

Table 6.—Number of blossoms and yield of Acala 1517C cotton in Experiment 2.

Treatment	No. of Blooms[a]		Yield of Cotton[b]		
	7/17	7/25	9/25	11/12	Total
Bayer 19639 50% Carbon	6.1	9.9	0.7	0.6	1.3
Bayer 19639 50% Emulsion	3.2*	9.8	0.5*	0.6	1.1
Thimet 44D	3.7	10.9	0.7	0.7*	1.4
Untreated	9.8	13.2	0.8	0.6	1.4
L.S.D. at 5% level[c]	6.04	4.81	0.17	0.14	0.46
at 1% level	11.09	8.83	0.32	0.26	0.46

[a] Number of blooms per 10 plants.
[b] Pounds of cotton per 10 feet of row.
[c] Based on four samples per plot.
* Significant at the 5% level.

differences in yield between the check and treatment plots were found (table 6).

CONCLUSIONS.—From the data obtained in Experiment 1, it is apparent that although the Bayer 19639 and Thimet treatments reduced the seedling stand of Pima S-1 and the Thimet-treated plots retarded blossoming and reduced growth at least through June 26, no reduction in yield resulted from these treatments.

Data from Experiment 2, where Acala 1517C cotton was tested, show slightly different reactions to the treatment. The end result in yield of cotton in this experiment was similar to Experiment 1. Based on these experiments, it appears that Acala 1517C was slightly more sensitive to the two materials tested, but that this sensitivity was not expressed in a decrease in yield at the end of the season.

REFERENCES CITED

Hacskaylo, J. 1957. Growth and fruiting properties and carbohydrate, nitrogen and phosphorus levels of cotton plants as influenced by Thimet. Jour. Econ. Ent. **50**(3): 280–4.

Hacskaylo, J., and J. C. Clark. 1957. Thimet uptake by Pima S-1 cotton and its effect on seedling emergence. Jour. Econ. Ent. **50**(4): 501.

Parencia, Jr. C. R., J. W. Davis and C. B. Cowan. 1957. Further field tests with systemic insecticides employed as seed treatments. Jour. Econ. Ent. **50**(5): 614–7.

Reynolds, H. T., T. R. Fukuto, R. L. Metcalf and R. B. March. 1950. Seed treatment of field crops with systemic insecticides. Jour. Econ. Ent. **50**(5): 527–39.

House Fly Resistance to Diazinon[1]

ELTON J. HANSENS, *Rutgers University, New Brunswick, N. J.*

ABSTRACT

House flies from a barn where a dairyman reported failure of fly control and from four barns where excellent control was obtained in 1957 were found resistant to Diazinon in laboratory tests. Resistance was proportional to the amount of Diazinon used in the barns during the several years previously. The most resistant strain was reared in the laboratory for four generations without appreciable loss in resistance. From this strain two other strains were developed by selecting 4-day-old and 1-day-old flies each generation on deposits of Diazinon of 1.0 mg. per sq. in. and rearing from survivors. Resistance increased very rapidly when 1-day-old flies were used for the selection since these flies had not mated before selection and less rapidly with selection of 4-day-old flies. All three strains of flies showed resistance to Diazinon, DDT, methoxychlor, chlordane, dieldrin, lindane, parathion, malathion, dicapthon, Dow ET 57, Dipterex, and Bayer 21/199 in comparison with a non-resistant laboratory strain.

Diazinon was first used in New Jersey for control of house flies in dairy barns in 1952 (Hansens & Bartley 1953). This material has been tested extensively since that time in this state and elsewhere and is generally regarded as a very effective residual spray against house flies.

For several years the same barns have been sprayed with Diazinon as often as necessary for house fly control. Initially these treatments were made to test the efficacy of this new material. When it proved to be an outstanding fly-killer, tests were continued to determine whether fly populations resistant to Diazinon would appear by spraying the same barns with Diazinon year after year. In 1957 excellent control was obtained in all barns sprayed with Diazinon, but for the first time it was noted that possibly control was not quite as good as in previous years. Meanwhile Kieding (1956) had reported Diazinon resistance in Denmark.

In September, 1956, a dairyman near Toms River, New Jersey reported inadequate fly control with Diazinon. Flies were collected from this farm on September 23rd and in laboratory tests were exposed to Diazinon residues on plywood panels. Additional flies were collected on October 2nd and the test repeated. At the same time flies were collected from four other farms where Diazinon had been used. These farms and their history of application of organic phosphate insecticides are as follows:

[1] Paper of the Journal Series, New Jersey Agricultural Experiment Station, Rutgers University, the State University of New Jersey, Department of Entomology, New Brunswick. Accepted for publication February 13 1958.

TREATMENTS IN BARNS.—*McGuire Farm.*—In late July 1957 all barns including calf barns and bull barns were sprayed with 0.5% Diazinon. In September the main calf barn and part of the milking barns were sprayed with 2.5% Diazinon with poor results. Diazinon was used in 1956, but details of previous control procedures are unknown.

Barn 1—In 1953 three applications of 1% malathion plus sugar were made followed by an application of 1% Diazinon (WP) on August 31. From 1954 through 1957 1% Diazinon was applied three times and 0.5% was applied four times.

Barn 48.—In 1953 was treated with Diazinon-impregnated cords. For 1954, fly control measures are not known. Two applications of dicapthon were made in 1955, followed in 1956 and 1957 by three treatments with 0.5% Diazinon.

Barn 22.—From 1953 through 1957 1% Diazinon was applied five times and 0.5% was applied five times.

Barn 9.—From 1953 through 1957 1% Diazinon was applied eight times.

Flies were exposed on plywood panels treated with 1.0 mg. of Diazinon per square inch using the test procedure developed by Barber & Schmitt (1948). The residual was applied to the panel by diluting the proper weight of Diazinon emulsifiable solution with acetone and applying 5 ml. of the solution from a pipette taking care to get complete and even coverage. Flies were exposed for either 5 or 15 minutes and then retained for 24 hours when counts of mortality were made. Comparisons of the several field collected strains with the laboratory strain are shown in table 1 from which it is evident that there were Diazinon-resistant flies in all of the field collected strains. The degree of resistance was proportional to the number of applications of Diazinon with the highest resistance where Diazinon has been used for 5 years; *i.e.*, at Farm 9 (8 sprays of 1% Diazinon) and at Farm 22 (5 sprays of 1% and 5 sprays of 0.5% Diazinon). Resistance was greater at Farm 9 where the greater amount of actual Diazinon had been applied per square foot of surface area.

DIAZINON RESISTANT STRAINS.—Laboratory studies were instituted to determine how rapidly resistance would be increased by selection or lost without selection. Using the most resistant strain (those flies collected from Barn 9) three laboratory strains were established: (1) an unselected line (U) consisting of the flies collected from Barn 9; (2) a selected line (S) consisting of the flies from Barn 9 which survived exposure for 15 minutes on 1 mg. of Diazinon per square inch when tested as 4-day-old flies; and (3) a selected line (T) consisting of those 1-day-old flies of the S line in the first generation which survived exposure to residual deposits of Diazinon of 1.0 mg. per square inch for 15 minutes. In each subsequent generation selections were made by exposing 4-day-old flies of the S line and 1-day-old flies of the T line on 1.0 mg./sq. in. of Diazinon for 15 minutes and using survivors for the parents of the following generation of each line.

The progeny of these three lines were tested in each generation as 1-day-old flies and as 4-day-old flies. Data on the tests with 4-day-old flies are given in table 2 which show that with selection Diazinon resistance was greatly increased. In as few as three generations the T strain reached a level where only 3 or 4% of the flies were killed by a dosage (5 min. on 1.0 mg. per sq. in.) which kills 100% of the non-resistant laboratory strain. The selection of flies when they are 1-day-old individuals results in more rapid development of a resistant population because both males and females are being selected, whereas with 4-day-old flies the females have opportunity to mate with non-resistant males.

There was a drop in resistance in the unselected line in the first generation but an increase in resistance each generation thereafter. Flies exposed when 1-day-old were generally more susceptible than 4-day-old flies.

In addition to the data shown counts were taken of the flies knocked down at the end of the exposure period and 1, 2, and 4 hours after exposure began. These data show that the greatest number of flies were down between 15 minutes and 1 hour after exposure began. After an hour 5 to 10% of the total number of flies in the test revived and were normal when final counts were made after 24 hours.

RESISTANCE TO OTHER INSECTICIDES.—Extensive tests were conducted with the three strains in the fourth generation on 12 insecticides in comparison with our non-resistant laboratory strain (Wilson). Panels were prepared

Table 1.—Comparison of strains of flies collected on dairy farms previously sprayed with Diazinon.

STRAIN	PER CENT KILL 24 HOURS AFTER EXPOSURE ON DIAZINON 1.0 MG./SQ. IN. AS INDICATED		
	15 Min.	5 Min.	Unexposed
McGuire[a]	94.6%	85.1%	0%
Laboratory-reared (W)	100	100	0
McGuire	94.9	64.7	0
Barn 1	92.0	62.5	5.6
Barn 48	82.8	41.9	5.3
Barn 22	67.4	35.5	0
Barn 9	60.7	16.1	0
Laboratory-reared (W)	100	100	0

[a] Tested September 24. Flies collected at McGuire Farm September 23 and compared with laboratory-reared (Wilson) line.
All other flies collected October 2 in barns and compared with Wilson line.

Table 2.—Mortality of 4-day-old flies of Diazinon-resistant strains in laboratory tests on plywood panels treated with 1.0 mg. Diazinon per sq. in.

STRAIN	PER CENT MORTALITY		
	Unselected (U)	Selected (S) After Mating	Selected (T) Before Mating
	15-minute exposure		
Field collected	60.7[a]		
1st generation	87.2	77.7	
2nd generation	64.2	53.6	27.6[b]
3rd generation	55.9	42.3	8.6
4th generation	51.4	21.4	3.1
	5-minute exposure		
Field collected	16.1		
1st generation	46.6	27.2	
2nd generation	48.5	26.5	14.3
3rd generation	36.5	17.5	4.1

[a] Survivors were parents of "S" strain.
[b] Survivors of 1-day-old flies exposed for 15 minutes to 1.0 mg. Diazinon/sq. in. were used as parents of "T" strain.

Table 3.—Mortality of house flies of several strains in laboratory tests.

	Panel (Mg./Sq. In.)	Per Cent Mortality						
		Exposure (Minutes)	Strain W	Strain U	Strain S	Strain T	Exposure (Minutes)	Strain T
Diazinon (tech.)	1.0	15	100	50.0	33.3	2.9	60	29.4
Diazinon (emuls.)	1.0	15	100	53.1	27.8	1.4	60	34.6
Malathion	1.0	60	66.7	0	0	0		
Parathion	0.1	15	98.7	26.5	6.7	0	60	0
Dicapthon	0.1	15	61.8	2.4	0	0	60	0
Dow ET-57	0.1	15	98.8	11.4	7.9	0	60	5.8
Dipterex	1.0	60	100	14.3	10.4	1.0		
Bayer 21/199	1.0	15	100	34.1	8.9	2.6		
DDT	1.0	15	99.0	5.3	1.3	0	60	0
Methoxychlor	1.0	30	83.7	0	0	0	60	0
Dieldrin	1.0	15	100	95.3	82.4	50.0	60	89.3
Chlordane	1.0	15	100	42.9	40.0	22.5	60	50.0
Lindane	0.1	15	100	30.4	10.2	0	60	12.5

in the same fashion as in previous tests. Insecticides used were either pure or technical grade chemicals. These data are presented in table 3 which shows that the three resistant strains were resistant to all 12 insecticides and this resistance was directly related to the intensity of the selection each strain received on Diazinon. Thus, the "T" strain was most resistant. Perhaps the most surprising finding was the high degree of resistance of the "U" strain which had not been exposed to any insecticides for five generations. It is not known whether this strain showed such resistance in the field but it is reasonable to assume that it did show some resistance to chlorinated hydrocarbons since the fly population in Barn 9 (where the strain was collected) showed resistance to chlorinated hydrocarbons in 1951.

As far as we are aware this strain ("T") is more resistant to both chlorinated hydrocarbon and organic phosphate insecticides than any strain previously reported from field or laboratory. Even with an hour exposure, large numbers of flies survived. Further experiments are planned to determine the LD-50 of this strain on these insecticides and to study the physiology of these flies.

References Cited

Barber, G. W., and J. B. Schmitt. 1948. House flies resistant to DDT residual sprays. New Jersey Agric. Expt. Sta. Bull. **742.**

Hansens, E. J., and C. E. Bartley, 1953. Three new insecticides for house fly control in barns. Jour. Econ. Ent. **46**(2): 372–4.

Kieding, J. 1956. Resistance to organic phosphorus insecticides of the house fly. Science **123:** 1173–4.

Systemic Insecticidal Action on Bloodsucking Ectoparasites of Several Compounds Administered Orally to Rabbits[1]

A. D. Flynn and W. G. Eden,[2] *Agricultural Experiment Station, Alabama Polytechnic Institute, Auburn*

Abstract

Twenty-eight synthetic organic compounds were screened for systemic insecticidal activity by administering the compounds orally to rabbits and allowing blood-sucking arthropods to feed on the treated rabbits. All 28 compounds were tested against the bed bug, *Cimex lectularius* L. Five of the chemicals were tested against the Gulf Coast tick, *Amblyomma maculatum* Koch; the yellow fever mosquito, *Aedes aegypti* (L.); and the stable fly, *Stomoxys calcitrans* (L.).

At the highest dosages that failed to produce visible toxic symptoms in the rabbits, five of the compounds exhibited systemic activity against the bed bug. These compounds and the dosage rates to the rabbits in mg./kg. were: Bayer 18613, 1100; Bayer 19596, 625; Bayer 19994, 25; Bayer 13242, 25; and Bayer 17364, 7.

The five chemicals that showed systemic activity against the bed bug were tested against the Gulf Coast tick, yellow fever mosquito, and stable fly. None of them exhibited systemic action against the tick or mosquito. There were indications that the compounds had some systemic action against the stable fly, but the results were inconclusive.

Results of research within the past few years have rekindled the interest of entomologists in finding effective systemic insecticides for control of insect pests of livestock. Adkins *et al.* (1955) conducted experiments with several organophosphorus compounds to determine the systemic effects against arthropods feeding on rabbits. The test arthropods were the bed bug, *Cimex lectularius* L., and nymphs of the lone star tick, *Amblyomma ameri-*

[1] This investigation was supported in part by a research grant, PHS G-4065 (c) from Division of Research Grants of the National Institutes of Health, Public Health Service. Accepted for publication February 15, 1958.

[2] The authors wish to thank Mr. T. R. Adkins for assistance in dosing the animals used in this work.

canum L. Dipterex; O,O-dimethyl-O- (ethylmercapto) methyl thiophosphate (Bayer 18/178) and methyl demeton caused 100% mortality of bed bugs and ticks feeding on treated rabbits. Hexamethylphosphoramide caused 63% mortality of bed bugs and 100% mortality of ticks. Schradan had no effect on ticks, but killed 100% of the bed bugs.

Adkins *et al.* (1957) determined the systemic effect of Dipterex through rabbits on the bed bug and Gulf Coast tick, *Amblyomma maculatum* Koch. Dipterex killed 50% of the bed bugs at a dosage level of 14 mg./kg. The LD-50 to ticks through the rabbit was calculated to be 36 mg./kg.

The systemic effect of Bayer 21/199 sprays was studied by Brundrett *et al.* (1957) who found that emulsions and suspensions were almost 100% effective in preventing grubs from erupting in the backs of cattle. The average infestation in 14 treated animals was 0.2 grub per animal; the 11 untreated controls had maximum counts averaging 37.2 grubs per animal. Bayer 21/199 applied dermally in advance of the grub season functioned as a systemic insecticide to prevent subsequent damage.

Roth *et al.* (1957) tested Dow ET-57 as a systemic against cattle grubs, *Hypoderma lineatum* (DeVill) and *Hypoderma bovis* (L.), at doses of 10 to 200 mg./kg. This material was found to be effective against grubs present in the backs of animals as well as against the encystment of new larvae. Second-instar grubs were killed more rapidly and efficiently than third-instar grubs. The material failed to kill many of the grubs that were within 2 weeks of emergence. McGregor *et al.* (1957) found that Dow ET-57 exhibited systemic insecticidal action against the screwworm, *Callitroga hominivorax* (Cqrl.), and the stable fly, *Stomoxys calcitrans* (L.), when given orally or subcutaneously to guinea pigs to 100 mg./kg. Dow ET-57 administered orally at 100 mg./kg. to calves naturally infested with northern and common cattle grubs gave an estimated 97.2% control and completely prevented encystment of new larvae. The practicality of Dow ET-57 as a systemic insecticide for the control of the common cattle grub under field conditions has been demonstrated (Adkins 1957).

The search for compounds that exhibit systemic insecticidal activity in animals has been continued at the Alabama Station.

MATERIALS AND PROCEDURE.—Preliminary experiments were conducted to determine the relative toxicity of 28 compounds to rabbits and their systemic insecticidal activity through the rabbit to the bed bug. The compounds that exhibited systemic action against the bed bug were further tested against the yellow fever mosquito, *Aedes aegypti* (L.), the stable fly, and the Gulf Coast tick. The compounds used in the experiments were acetone; O,O-diethylphosphoroacetothioate, Bayer 13242; ethyl-chloroethyl-dichlorovinyl phosphate, Bayer 16574; O,O-diethyl-O- (carboethoxy-methylmercapto-ethyl) thionophosphate, Bayer 16900; vinylcarboxymethyl-dimethyl-dithio carbamic acid, Bayer 17243; N-succinyl-ethylthio-S- (O,O-diethyl-phosphorothioate), Bayer 17364; O,O-diethyl-O-phthaloximido thionophosphate, Bayer 17450; 3,4-dichlorobenzyl-p-chlorobenzene sulfonate, Bayer 18220; O,O-diethyl-S-p-chlorobenzyl-dithiophosphate, Bayer 18341; O,O-dimethyl-S-[(phenyl) (carboethoxy) methyl]-thiolphosphate, Bayer 18510; ethylglycolate-(O,O-diethyl-phosphorothioate), Bayer 18613; O-ethyl-O-isopropyl-O-phthaloximido thionophosphate, Bayer 18779; O,O-dimethyl-S-p-chlorobenzyl-thiol-thionophosphate, Bayer 18780; O,O-di-n-propyl-S-p-chlorobenzyl-thiol-thionophosphate, Bayer 18781; O,O-di-isopropyl-S-p-chlorobenzyl-thiol-thionophosphate, Bayer 18786; O,O-diethyl-0-2-diethylaminoethyl-thionophosphate, Bayer 18935; mixed anhydride of O,O-diethyl-thionophosphoric acid and benzenesulfonic acid, Bayer 19285; S-aceto-ethyl-carbamate-O,O-diethyl-phosphorodithioate, Bayer 19596; Bayer 19639; O,O-diethyl-S-[(phenyl) (cyano) methyl] thiol-thionophosphate, Bayer 19641; O,O-diethyl-O-2-pyridino- (2)-ethyl-thionophosphate, Bayer 19994; mixed anhydride of O,O-diethyl-thionophosphoric acid and bis-dimethylamidophosphoric acid, Bayer 20172; O-ethyl-S- (ethylmercaptomethyl)-dithiocarbonic acid, Bayer 20743; Bayer 28/63; demeton; DDVP; Hercules AC-528; and Thimet.

Most of the compounds used in the experiments were not water soluble. They were given to the rabbits as acetone solutions and acetone-water emulsions. Those that were water-soluble were given as water solutions. All compounds were formulated to contain 100 mg./ml. when given to the rabbits. The chemicals were administered through a stomach tube (Bardam No. 12 Fr. urinary catheter) to domestic rabbits, which were restrained in specially constructed stocks.

Mammalian toxicities were determined by a modification of procedure described by Adkins *et al.* (1955). The approximate highest sublethal dose that could be given to the rabbits without causing severe visible toxic symptoms was determined. The toxicities of most of the compounds were not known. Therefore, a starting dose was arbitrarily selected. If the first dose killed the rabbit, successively lower doses were given until one failed to kill. Conversely, if the first dose caused no visible toxic symptoms, successively larger ones were administered until an animal was killed. If the highest dose that failed to kill the animal did not kill all of the test arthropods, the chemical was eliminated from further testing. No rabbit was used twice in any experiment.

Stable flies, female yellow fever mosquitoes, fifth-instar bed bugs, and nymphs of the Gulf Coast tick were placed on the treated rabbits to feed. All of the test insects were laboratory reared. A different animal was used for each test arthropod to prevent injury to the rabbit from excessive handling.

Eight to 31 fifth-instar bed bugs, depending on the number available, were allowed to feed to repletion on each rabbit 1 hour after the rabbit had received a dose of the chemical. The stable flies and yellow fever mosquitoes were handled in a similar manner, except the male mosquitoes were removed from the test chambers to prevent them from interfering with the females during the feeding operation. From 7 to 100 nymphs of the Gulf Coast tick were allowed to attach to the rabbits. These animals were dosed 2 days after the ticks were attached. Four to 5 days later the ticks had engorged and dropped. Mortality counts were taken on all lots 24 hours after dosing the animals and again when all of the ticks in the check had

Table 1.—Summary of results of toxicity experiments with rabbits that received oral doses of various insecticides.

Insecticides	No. of Rabbits	Dose to Rabbits (Mg./Kg.) Highest Sublethal	Lowest Lethal
Acetone	4	5 ml./kg.	6 ml./kg.
Bayer 13242	6	25	35
Bayer 16574	7	10	20
Bayer 16900	4	50	75
Bayer 17243	5	325	350
Bayer 17364	7	7	10
Bayer 17450	6	330	400
Bayer 18220	6	260	300
Bayer 18341	6	200	300
Bayer 18510	4	50	75
Bayer 18613	7	1100[a]	1100[a]
Bayer 18779	5	100	135
Bayer 18780	6	230	275
Bayer 18781	6	350	400
Bayer 18786	5	325	350
Bayer 18935	9	900	1000
Bayer 19285	5	5	10
Bayer 19596	10	625	650
Bayer 19639	7	2	5
Bayer 19641	5	25	40
Bayer 19994	5	25	40
Bayer 20172	4	15	25
Bayer 20743	6	200	250
Bayer 28/63	3	25	50
Demeton	3	1	2
DDVP	12	25	30
Hercules AC-528	5	25	50
Thimet	8	2	5

[a] These figures were determined from different shipments of 18613.

molted. After feeding, the test arthropods were kept in holding chambers in a constant temperature box maintained at 80°F. A piece of cotton moistened with a solution of sucrose was given to the stable flies and the yellow fever mosquitoes until the final counts had been made.

RESULTS.—Results of the toxicity experiments on the rabbits are presented in table 1. Of the materials tested, Bayer 18613 was the least toxic to the rabbits and demeton was the most toxic. The maximum sublethal dose of demeton was 1 mg./kg, and that of Bayer 18613 was 1100 mg./kg.

The data on systemic action of the compounds against the bed bug are presented in table 2. Five of the compounds exhibited systemic activity. Bayer 19596, Bayer 13242, and Bayer 17364 were all highly systemic, resulting in 100% mortality of the bed bugs 24 hours after feeding on the dosed rabbits. The respective dosages of the toxicants to the rabbits of 625, 25, and 7 are, of course, not indicative of the relative systemic insecticidal activity of the compounds. These were the highest sublethal dosages to the rabbits. Bayer 19994 and 18613 also showed systemic activity resulting in 90% kill of the bed bugs 6 days after they fed on the rabbits.

None of the five chemicals showed systemic action against the yellow fever mosquito or the Gulf Coast tick.

Table 2.—Mortality of fifth-instar bed bugs that fed on rabbits dosed orally with insecticides.

Insecticide	Dose to Rabbits (Mg./Kg.)	No. Bed Bugs Used	Per Cent Mortality After Feeding 24 Hours	6 Days
Check	0	10	0	0
Acetone	5 ml./kg.	16	0	0
Bayer 13242	25	20	100	0
Bayer 16574	10	20	0	0
Bayer 16900	50	10	0	0
Bayer 17243	325	20	0	0
Bayer 17364	7	20	100	0
Bayer 17450	330	15	0	0
Bayer 18220	260	12	0	0
Bayer 18341	200	10	0	0
Bayer 18510	50	10	0	0
Bayer 18613	1100	10	20	90
Bayer 18779	100	20	0	0
Bayer 18780	230	20	0	0
Bayer 18781	350	10	0	0
Bayer 18786	325	20	0	0
Bayer 18935	900	15	0	0
Bayer 19285	5	20	0	0
Bayer 19596	625	20	100	0
Bayer 19639	2	20	0	0
Bayer 19641	25	10	0	0
Bayer 19994	25	10	90	90
Bayer 20172	15	10	0	0
Bayer 20743	200	20	0	0
Bayer 28/63	25	10	0	0
Demeton	1	10	0	0
DDVP	25	10	0	0
Hercules AC-528	25	20	0	0
Thimet	2	20	0	0

There were some indications that the compounds showed systemic activity against the stable fly, but the results were inconclusive.

REFERENCES CITED

Adkins, T. R., W. L. Sowell, and F. S. Arant. 1955. Systemic effect of selected chemicals on the bed bug and lone star tick when administered to rabbits. Jour. Econ. Ent. 48(2): 139–41.

Adkins, T. R., and F. S. Arant. 1957. Systemic effect of Dipterex on the bed bug and Gulf Coast tick when administered to rabbits. Jour. Econ. Ent. 50(2): 166–8.

Adkins, T. R. 1957. Field evaluations of Dow ET-57 as a systemic insecticide for the control of the common cattle grub in Alabama. Jour. Econ. Ent. 50(4): 474–6.

Brundrett, H. M., W. S. McGregor, and R. C. Bushland. 1957. Systemic cattle grub control with Bayer 21/199 sprays. Agric. Chemicals 12(6): 36.

McGregor, W. S., and R. C. Bushland. 1957. Tests with Dow ET-57 against two species of cattle grubs. Jour. Econ. Ent. 50(3): 246–9.

Roth, A. R., and Gaines W. Eddy. 1957. Tests with Dow ET-57 against cattle grubs in Oregon. Jour. Econ. Ent. 50(3): 244–6.

Resistance to Boll Weevil in Mixed Populations o Resistant and Susceptible Cotton Plants[1]

W. H. Wessling,[2] *Genetics Faculty, North Carolina State College, Raleigh*

Abstract

The reliability of resistance to attack by boll weevil (*Anthonomus grandis* Boh.) conferred by the mutant gene, H_2, was tested under different environmental conditions, *i.e.*, in different proportions of the mutant strain, Pilose, and the susceptible variety, All-In-One, or in the absence of susceptible plants. In order to carry out this test it was necessary to isolate individual plots from one another using corn as a barrier crop, and to provide a uniform infestation of boll weevils. This method proved satisfactory. The degree of resistance was measured inversely by the proportion of squares with egg-laying punctures. The mutant strain, Pilose, showed a significantly lower proportion of egg-laying punctures throughout the investigation. A slight but consistent increase in number of egg-laying punctures with increase in the number of susceptible plants in the mixture was noticed in the resistant strain, Pilose, as well as in the susceptible variety, All-In-One.

Different sources of resistance to the boll weevil (*Anthonomus grandis* Boh.) in strains of Upland cotton were pointed out by Stephens (1957). He also reviewed the literature pertinent to the problem. In a second paper in this series, Wannamaker (1957) reported preliminary investigations with two mutant genes, H_1 and H_2, either of which produces a higher degree of plant hairiness than normally found in commercial Upland cottons. He found that the number of egg-laying punctures was reduced to the same extent in strains carrying either of these mutant genes in homozygous condition. Wannamaker's experiment included a large number of susceptible types in addition to the resistant mutant strains, and he raised the question whether the resistance shown by the mutant strains would be reduced or increased under conditions in which no susceptible strains were available for oviposition in the immediate neighborhood. Also insecticides were applied during the course of his experiment, and it was possible that part of the observed resistance might have been due to the fact that hairy plants tend to hold insecticidal dusts better than non-hairy plants, as pointed out earlier by Loftin (1946).

It is the purpose of this paper to present the results obtained from an experiment designed to test whether or not the degree of resistance expressed by a resistant strain would be affected by the relative proportions of resistant and susceptible plants in a mixed population, and in the absence of any form of insecticidal control.

Materials and Methods.—(1) *Plant material.*—The resistant mutant strain and the susceptible commercial variety used in the experiment may be described as follows: The mutant strain, Pilose, carries the partially dominant gene, H_2, which in the homozygous state confers a dense pubescence covering stems, leaves, bracts, flower-buds and bolls. The susceptible variety, All-In-One, carries the "normal" allele, h_2, in homozygous condition. It is an Upland cotton variety grown in the southeastern United States and has the small amount of hairiness inherent in all commercial Upland cotton varieties. For purposes of isolation (see below) the corn variety, N. C. 46, was used. This variety was chosen because of its compact growth and tillering capacity.

(2) *Cultural practices.*—The cotton seeds were planted in early May, in paper cups in the greenhouse, grown for 2 to 3 weeks until the first true leaf developed, hardened for another 2 weeks and then transplanted to the experimental field. This was located at the Method Horticultural Station near Raleigh, N. C., where there was no record of recent cotton cultivation nor was any cotton growing within approximately 2 miles. The experimental field was first planted with corn and the plots were cut out when the cotton was transplanted. Row width for both cotton and corn was standardized at 42 inches, the corn being spaced at 2 feet in the row with three plants per hill. The cotton plants were planted individually 1 foot apart in the row. Normal cultural practices were observed during the course of the experiment.

(3) *Experimental design.*—A randomized complete block design was used, each treatment being replicated five times. The four treatments consisted of

	Pilose (resistant)	*All-In-One (susceptible)*
(a)	100%	0%
(b)	75%	25%
(c)	50%	50%
(d)	25%	75%

Each plot contained six rows, each row 10 plants. The two outer rows and the end plants of the four middle rows served as borders, so that the experimental material in each plot consisted of four rows of eight plants each. In the case of the mixed treatments (b, c, and d) the resistant and susceptible plants were arranged systematically in the plot. This was done in such a manner that each susceptible plant was surrounded by resistant plants in Treatment (b) and vice versa in Treatment (d). Resistant and susceptible plants were alternated in Treatment (c). This systematic arrangement was preferred to randomization within each plot because it provided more uniform conditions for infestation (*i.e.*, chance "pockets" of resistant or susceptible plants were avoided).

In order to compare differences between treatments it was necessary to provide a uniform initial infestation of weevils in each plot and to prevent migration from plot to plot until the termination of the experiment. For this reason each cotton plot was isolated from its neighbors by

[1] Contribution from the North Carolina Agricultural Experiment Station. Published with the approval of the Director as Paper No. 884 of the Journal Series. Part of a thesis presented in partial requirement for the Ph.D. degree. Accepted for publication February 24, 1958.

[2] Present address: c/o Schreiber & Sohne, 237 Boeckler Str., Braunschweig, Germany.

a corn plot of equal area (21 ft.×10 ft.) on every side. The whole experimental area was surrounded by four rows of corn to protect it from possible immigrant weevils. As will be shown later, N. C. 46 served these isolation purposes very well.

To make sure that no natural infestation was present at the beginning of the experiment, the area was dusted twice with toxaphene. After the second application 2 weeks were allowed to elapse in order to eliminate possible residual effects. Infested squares were then collected in South Carolina and approximately equal numbers of them were distributed in each plot, in such a way that they fell close to the plants along every row. Light rain fell a short time after the squares were distributed, and the cool moist conditions favored the development and emergence of the weevils. Judging by the numbers of squares distributed it was estimated that at least one adult weevil was provided for every cotton plant in the test. During the course of the experiment no insecticides were applied. The experiment, therefore, can be considered a very severe test of resistance to boll weevil attack.

(4) *Method of scoring.*—Six plants of each strain were selected at random in each plot and scored. In the plots containing 100% of resistant plants, two alternate samples of six plants each were scored. Three days after the majority of the weevils had left the squares, scoring was started. All selected plants were scored twice a week for 4 consecutive weeks (*i.e.*, until the next generation of weevils appeared in the middle of August). Scoring was stopped then because it was considered that the bolls which were set after late August would hardly add much to the total yield, and therefore to the information needed.

Every square on each selected plant was scored by close examination for feeding and egg-laying punctures. As soon as the observation was made an "O.K. Paper Fastener" was used to mark the square. The size of the square determined if it was to be scored at a certain date or later. As soon as a flower bud opened, the square next to it on the same sympodium was chosen for scoring. This procedure held as a general rule for the resistant strain, Pilose. It also held for the susceptible variety, All-In-One, as long as relatively few squares were damaged, *i.e.*, at the beginning of the experiment. As soon as squares became rare, however, it was observed that even very small squares were punctured and fell off very quickly. The procedure was then changed and smaller squares were scored as soon as they were punctured so as not to lose information. These were re-examined later if they remained on the plant and the data were corrected if changes had occurred in the meantime. It should be noted at this point that both strains started flowering at the same time and set bolls in essentially the same manner.

RESULTS.—All counts of squares showing no punctures, feeding punctures and egg-laying punctures were converted first to percentages. Because the percentages resulted from fewer than 100 counts in most cases, and ranged from less than 20% to more than 80%, they were transformed to angles (angle = arc sin $\sqrt{\text{percentage}}$) for statistical analysis (Fisher & Yates 1943).

In figure 1 the proportions of squares with feeding, egg-laying and no punctures in the four treatments are presented. The figure shows clearly that the differences between treatments and between strains within each treatment lie in the proportions of egg-laying punctures. No significant differences could be found between any two means of feeding punctures. Since the proportion of feeding punctures tended to stay constant, reduction in proportion of egg-laying punctures was directly proportional to increase in proportion of unpunctured squares.

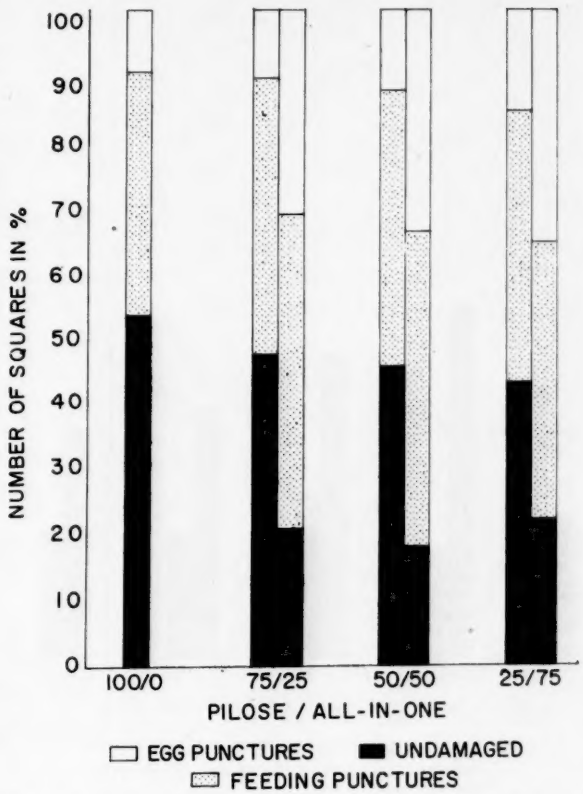


FIG. 1. Comparisons of relative damage due to boll weevil injury in different mixtures of Pilose and All-In-One.

To obtain further information on the effects of resistance, bolls were counted 2 weeks after the scoring of squares was discontinued (fig. 2). The writer is aware that these counts may not be directly related to the number of unpunctured squares, since other factors like disease, bollworm (*Heliothis zea* (Boddie)), or climatic factors can cause loss of bolls. The observation adds, however, to the information which would be supplied otherwise by a yield test. A comparison of figures 1 and 2 shows that the average number of bolls set per plot in the different mixtures agrees very well with the observations made on the squares.

Since the chief differences between the two strains as shown in figure 1 were clearly associated with the relative proportions of egg-laying punctures, the latter were subjected to statistical analysis. Table 1 shows the mean percentages of egg-laying punctures and their corresponding angular transformations which were obtained in the four treatments. Differences between the resistant and susceptible components in the three mixed treatments are analyzed by t-tests shown in the table. All the t-values

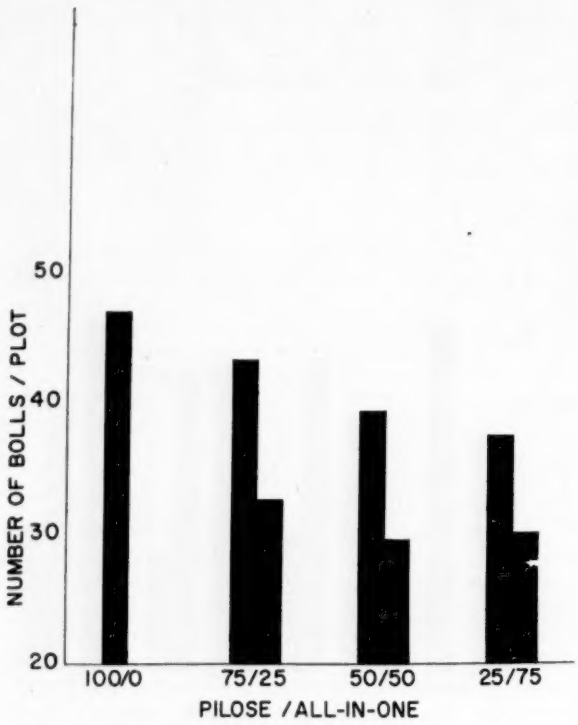


FIG. 2.—The number of undamaged bolls per plot in different mixtures of Pilose and All-In-One.

have probabilities lower than 1% which shows that no matter how high or low is the proportion of the resistant Pilose plants in the mixture, the difference between the two strains under consideration remains highly significant. Further examination of the data in table 1 shows that the mean number of egg-laying punctures in the resistant Pilose plants tends to be reduced as the proportion of Pilose plants in the mixture becomes greater. That this apparent trend is a real one is shown by the analysis of variance presented in table 2, in which the differences in egg-laying punctures between Pilose plants in the various mixtures are seen to be significant. This means that the resistance of the Pilose strain, as measured inversely by the proportion of egg-laying punctures, becomes greater as the proportion of Pilose plants in the mixture increases.

The fact that the Pilose plants in the various mixtures differed signficantly in the proportion of egg-laying punctures in itself indicates that the method of isolation had well served its intended purpose. Otherwise no noticeable difference between treatments, except those caused by chance, would have been expected. Also the fact that the interaction item, "days × replicates," in the same table is highly significant adds to the evidence that the isolation was at least partially effective.

Table 1.—Comparisons of percentages of squares with egg-laying punctures obtained in different mixtures of Pilose (resistant) and All-In-One (susceptible) strains of Upland cotton.

MIXTURE	SAMPLE SIZE	OBSERVED MEAN PER CENT	MEANS OF ANGLES	DIFFERENCES	t-VALUES	P
Pilose 100%	1563	6.71	15.05	—	—	—
Pilose 75%	810	6.55	14.83	14.59	5.23	<.01
All-In-One 25%	937	24.10	29.42			
Pilose 50%	761	9.46	17.54	12.06	7.09	<.01
All-In-One 50%	903	24.40	29.60			
Pilose 25%	738	11.94	20.06	12.21	5.05	<.01
All-In-One 75%	778	28.70	32.27			

Table 2.—Analysis of variance of transformed percentages of egg-laying punctures in the mutant strain Pilose.

SOURCE	DEGREES OF FREEDOM	MEAN SQUARES	F	P
Mixtures	3	251.16	3.55	.05–.01
Replicates	4	147.86	2.09	n.s.>.05
Days	7	1994.51	28.17	<.01
Days×Mixts.	21	87.63	1.24	n.s.>.05
Days×Reps.	28	163.22	2.31	<.01
Reps.×Mixts.	12	93.76	1.32	n.s.>.05
Days×Reps.×Mixts.	84	70.81		
Total	159			

These data as a whole provide answers to the main problems which the experiment was designed to solve, *i.e.*, they show (1) that the Pilose strain is consistently more resistant than the All-In-One variety over a range of mixtures (2) that there is a tendency for the resistance of Pilose to become greater as its proportion in the mixture is increased and (3) that its resistance is independent of the application of insecticides. There remains for consideration the question of what stage in the life history of the plant and at what particular part of the insect's life cycle does resistance become effective. A certain amount of information concerning this question can be obtained by examination of table 3 and figure 3. Table 3 shows the proportion of squares with egg-laying punctures, with feeding punctures and without punctures, in per cent, at successive dates of scoring. When we examine the proportion of squares with egg-laying punctures we find that in the resistant strain, Pilose, the proportion stays low in the early counts and increases slowly in the treatments which contain a higher number of resistant plants—more rapidly in the treatments with a lower number of resistant plants. The same tendency is found to a somewhat lesser extent in the susceptible variety, All-In-One. Clearly the presence of Pilose in the mixtures is associated with a delay in egg-laying.

This effect is shown in more striking fashion when the averaged egg-laying sequences in the two strains are compared. In figure 3 the data shown in table 3 are averaged over all mixtures for each strain separately and presented graphically. In the susceptible variety there is a rapid increase in the proportion of egg-laying punctures as soon as the maturation feeding period is passed, *i.e.*, approximately 1 week after the emergence of the boll weevils from the squares (Hunter & Pierce 1912). In the resistant strain the proportion of egg-laying punctures increases much more slowly and never reaches the magnitude attained by the susceptible variety. Since the percentages are based on a comparable number of squares counted in each case (see table 1, "Sample size") it may be concluded

Table 3.—Percentages of squares with no punctures (0), with feeding punctures (FP), and with egg-laying punctures (EP), at successive dates of scoring.

	PILOSE			ALL-IN-ONE		
DATE	0	FP	EP	0	FP	EP
		100%			*0%*	
7/16	56.95	43.05	0.00	—	—	—
7/21	40.21	56.08	3.70	—	—	—
7/24	59.62	31.73	8.65	—	—	—
7/27	74.78	20.00	6.96	—	—	—
7/31	74.86	13.14	12.00	—	—	—
8/4	71.23	17.35	11.42	—	—	—
8/7	36.64	49.14	14.22	—	—	—
8/10	31.03	50.43	18.53	—	—	—
		75%			*25%*	
7/16	40.36	59.64	0.00	28.47	71.53	0.00
7/21	50.00	47.41	2.59	18.48	68.48	13.04
7/24	50.00	42.11	7.89	39.62	43.40	16.98
7/27	60.32	21.75	7.93	42.65	22.06	35.29
7/31	68.18	27.27	4.55	45.00	18.75	36.25
8/4	65.09	25.49	7.84	28.93	31.40	39.67
8/7	28.10	47.93	23.97	6.83	50.24	42.93
8/10	32.43	36.94	30.63	3.95	49.15	46.89
		50%			*50%*	
7/16	34.57	65.43	0.00	21.74	78.26	0.00
7/21	43.48	51.30	5.22	23.71	57.73	18.56
7/24	60.00	35.56	4.44	58.53	34.15	7.32
7/27	76.56	7.81	15.63	36.17	23.40	40.43
7/31	64.84	23.08	12.09	33.70	19.56	46.74
8/4	61.25	25.00	13.75	27.88	22.12	50.00
8/7	20.41	51.02	28.57	9.09	57.81	37.50
8/10	29.47	46.32	24.21	10.99	44.51	54.40
		25%			*75%*	
7/16	38.92	61.08	0.00	31.78	68.22	0.00
7/21	46.67	48.89	4.44	32.82	55.90	11.28
7/24	48.08	28.85	23.08	30.30	51.52	18.18
7/27	58.20	20.90	20.90	31.41	25.71	42.86
7/31	62.86	22.86	14.28	53.42	19.18	27.40
8/4	56.10	20.73	23.17	25.56	28.89	45.56
8/7	24.76	44.76	30.48	7.33	34.67	58.00
8/10	20.59	49.02	30.39	4.14	39.05	56.81

that the susceptible variety was at a progressively greater disadvantage during the period of study.

DISCUSSION.—From the results obtained in this experiment it was concluded that the chief difference between the resistant and susceptible strains was in the proportion of squares with egg-laying punctures. In all mixtures the resistant strain had a significantly lower proportion of egg-laying punctures than the susceptible variety, which showed that this form of resistance was consistent. Further, the degree of resistance exhibited by the resistant strain increased as the proportion of susceptible plants in the mixture was reduced. One might expect that boll weevils would not easily switch to a resistant strain of this type, if susceptible varieties were no longer available.

Successive counts of egg-laying punctures which were made over a 4-week period showed that the proportion of squares with egg-laying punctures increased very rapidly in the susceptible variety, while in the resistant strain egg-laying was delayed at the outset and increased relatively slowly. These different patterns of response in the resistant and susceptible strains have an important two-fold implication for the potential yield of the crop. Under normal conditions of growth it is the earliest bolls set which contribute most to yield of the Upland cotton plant. Consequently potential yield will be considerably

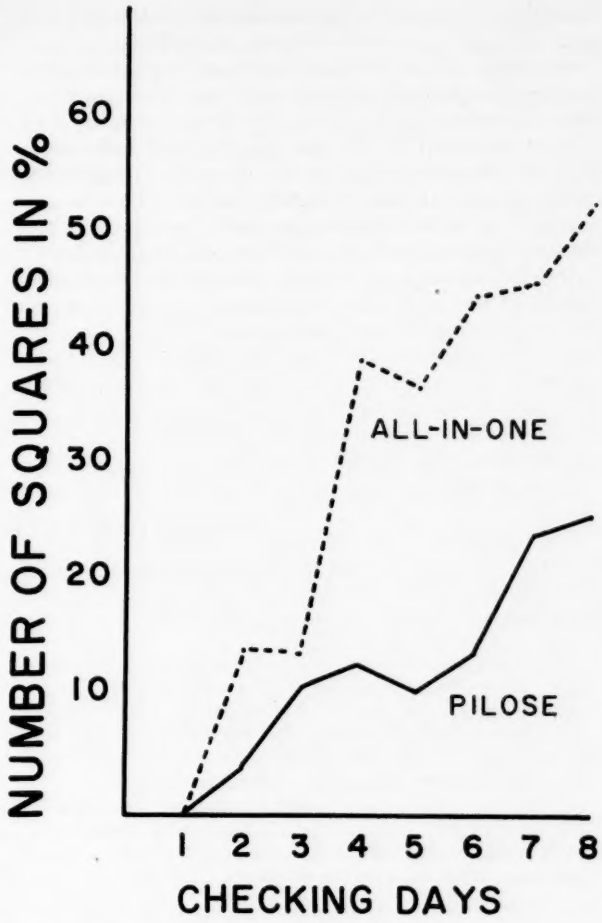


FIG. 3.—The percentages of squares with egg-laying punctures on successive checking days in Pilose and All-In-One. The percentages are averaged over all treatments.

increased if heavy loss of squares resulting from early boll weevil attack is delayed for a certain time so that more squares can develop early in the season. Secondly, a delay in egg-laying will also delay the emergence of the following generation of the insect and therefore allow a longer period for the setting of the main crop. In both these respects the resistant strain, Pilose, was at a considerable advantage as compared with the susceptible variety, All-In-One.

Another problem has attracted the writer's attention from a practical as well as a theoretical point of view. How does the presence of a large number of susceptible plants affect the success of relatively few, moderately resistant plants in the same population? It was found that the effectiveness of resistance decreases consistently with an increasing proportion of susceptible plants. It can be assumed that eventually when the number of resistant plants becomes very low, the effect of resistance will be hardly detectable in a large breeding population. This can be expected especially for plant populations where insect infestation is high and the initial degree of resistance which may arise by mutation in a single plant is relatively

low. It does not apply to populations in which a high degree of resistance or even immunity is available.

A moderate degree of insect resistance may then evolve in a natural plant population under two circumstances: when insect infestation is relatively low or when the proportion of mutants in the plant population is sufficiently high that the resistant plants are an essential proportion of the population and, therefore, have a sufficient advantage. In a breeding program where only a moderate degree of resistance is to be expected this should be borne in mind and a very careful study of each individual plant should be made in a large population to detect the single or few plants which may promise success.

References Cited

Fisher, R. A., and F. Yates. 1943. Statistical tables for biological, agricultural, and medical research. London, Oliver and Boyd. 98 pp.

Hunter, W. D., and W. D. Pierce. 1912. The Mexican cotton boll weevil. U. S. Dept. Agric. Ent. Bull. **114.**

Loftin, U. C. 1946. Living with the boll weevil for fifty years. Smithsonian Inst. Rept. Publ. **3827.**

Stephens, S. G. 1957. Sources of resistance of cotton strains to the boll weevil and their possible utilization. Jour. Econ. Ent. **50**(4): 415–8.

Wannamaker, W. K. 1957. The effect of plant hairiness of cotton strains on boll weevil attack. Jour. Econ. Ent. **50**(4): 418–23.

Aestivation of the Alfalfa Weevil[1]

G. R. Manglitz,[2] *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*

Abstract

Since the alfalfa weevil (*Hypera postica* (Gyll.)) was discovered in the Eastern States, there have been conflicting reports on the number of generations per year and its summer activity in general. Extensive sampling during the summer showed that most of the adults were in surface litter and soil under trees, hedge rows, and fence rows bordering alfalfa fields. The exodus from the fields may be in response to high temperature. There is probably not more than one generation in a season. The small numbers of immature stages sometimes found in the summer appear to be the progeny of overwintered adults.

Several workers have made observations on the biology of the alfalfa weevil *Hypera postica* (Gyll.) since its discovery in the eastern United States. According to Milliron & MacCreary (1955), a second generation is produced during the summer. App (1954) found indications of a partial second generation in the fall. Later Manglitz & App (1957) observed that late-fall larvae seldom reach maturity. They believe that there is but one generation and that after a period of vigorous feeding, the newly emerged adults leave the alfalfa fields and do not return until fall.

The study herein reported, conducted in Maryland during the summer of 1957, was made primarily to answer the question as to where the alfalfa weevil adults go during the summer. This exodus from alfalfa fields may be associated with the inability of the female to lay eggs for several months after reaching the adult stage. This fact has been noted by many workers, among them Snow (1928) who by means of dissections, demonstrated that in Utah, although some females are capable of egg deposition in the fall, many pass the winter before ovulation is completed.

Procedure.—The general plan of this study was to sample the areas surrounding a number of fields. Samples were taken by means of a 1-square-foot steel frame. Surface trash and/or plant material was removed from inside the frame, and then soil was removed to the depth of 1 inch. Plant material was examined visually, and trash samples were generally run in a modified Berlese funnel. The soil was screened over hardware cloth, and then washed on a finer mesh screen.

Six alfalfa fields were selected that were surrounded by areas typical of central Maryland. These fields ranged in age from the second to the fourth cropping year and in size from 3 to 25 acres. The design of sampling and number of square-foot samples taken per field varied with the types of vegetation and cropping systems.

Field No. 1 located near Beltsville, Prince George's County, was surrounded by dense woods. Eleven series of seven samples each were taken around the field. The first sample in each series was taken just inside the edge of the alfalfa, the second sample into the woods 2 feet from the first, and the third, also in the woods, at 4 feet from the second. The following intervals were 8, 16, 32, and 64 feet, making the last sample 126 feet into the woods from the edge of the field. In addition, 20 samples were taken near the center of the field.

In an effort to establish reasons for the adult weevils leaving the field, a hygrothermograph was placed near the center of the field in a screened enclosure with a solid roof. The instrument was on a wooden platform about 3 inches above the ground. Another hygrothermograph, which had been synchronized for several days with the first one, was placed in an identical cage several feet from the field border into the woods.

Field No. 2 was also located near Beltsville. Fields No. 3 to 6 were in Howard and Anne Arundel Counties. None were surrounded by woods like the first field, and various conditions were represented.

In addition to the field samples, insectary observations were made on adults of known ages that were held in 1-gallon plastic containers with cheesecloth tops. The amount of post-emergence feeding was varied. Oviposi-

[1] Accepted for publication February 24, 1958. Presented before the meeting of the Eastern Branch of the Entomological Society of America, New York, N. Y., November 25–26, 1957.

[2] W. P. Dodson and R. A. Berry, Jr., biological aids, assisted in this study.

Table 1.—A comparison of temperatures (°F) between woods near field edge and center of field No. 1, Beltsville, Md., 1957.

DATE	CENTER (A)	WOODS (B)	A–B
July 24	73.0	69.5	+3.5
25	72.5	70.0	+2.5
26	74.5	71.5	+3.0
27	76.5	76.0	+0.5
28	78.5	73.5	+5.0
29	79.5	75.0	+4.5
30	81.0	78.0	+3.0
31	80.0	77.5	+2.5
August 1	78.5	77.5	+1.0
2	79.5	74.5	+5.0
3	85.5	81.5	+4.0
4	74.5	69.0	+5.5
5	72.0	72.5	−0.5
Mean	77.4	74.3	+3.1
L.S.D. at 1% level		1.32	

tion observations were made three times a week and survival counts once a week.

RESULTS.—The samples taken in the woods surrounding field No. 1 showed that the greatest mean number of weevils, 1.18 per square foot, was found 2 feet from the edge of the field. A mean of 0.73 per square foot was found at the 62-foot distance. Less than 0.30 weevil per square foot was found at the remaining distances and at 126 feet no weevils were found. No weevils were found in the center of the field.

The temperature records at two locations in field No. 1 for a 13-day period are presented in table 1. The mean temperature in the center of the field was 3.1° F. higher than that several feet into the woods. Since this difference is highly significant, it is possible that the weevils spend the summer out of the fields to escape the higher temperature present there. It is unfortunate that the instruments used failed to record relative humidity properly, because Sweetman & Wedemeyer (1933) indicate that under controlled conditions relative humidities below 40% with temperatures 27° C. (80.6° F.) or higher were very destructive to the adults.

The counts in all the fields are summarized in table 2. It may be seen, although observations on this point are limited, that no weevils were found in unprotected places such as grain stubble and corn fields, and few were found in other open areas. In only one field (No. 6) adults were found in the center of the field, and it had more surface trash than usual because one recent hay cutting had been allowed to remain and the field had received a heavy application of manure. This field was also surrounded for some distance by corn, which, with frequent cultivations, apparently affords poorer protection than the alfalfa.

In order to determine whether there were any weevils below the 1-inch level, field No. 2 was revisited, and ten additional 1-square-foot samples were taken at random from a hedge row where the greatest concentration of adults had been observed. These samples were separated into four layers—surface trash and litter, the top inch, the second inch, and the third inch of soil. The mean number of adults per square foot in the trash and litter samples was 2.4, which was statistically significant from the soil means except the one for the first inch of soil, which was 1.6 adults per square foot. The means for the deeper samples were 0.5 in the second inch and 0.2 in the third inch. There was no significance between the means for the soil samples.

Slight midsummer buildups of larvae, as mentioned by Milliron & MacCreary (1955) and for which Manglitz & App (1957) give detailed records over a 4-year period, apparently are responsible for the reports of a second generation. The summer larval activity was not observed in 1954 and did not occur during the 1957 season. It was at first thought that these midsummer larvae might be the progeny of very early maturing adults. However, in 1957 weevil activity began considerably earlier than normal, hatching being observed on March 12, whereas the next earliest record was April 2, 1954.

Several hundred adults emerging in early May were held in cages during the summer, but no oviposition was observed. Overwintered adults were also held during this period and, although several survived until mid-July and one until late August, none deposited eggs during the last month or so of life. Although it cannot be stated definitely that summer larvae result from eggs from overwintered adults, it is shown that overwintering adults can live long enough to be responsible for such larvae. Also it is possible that in seasons when oviposition begins early the egg-laying capacity of overwintered adults is depleted before midsummer. The two seasons in which few summer larvae were found were the earliest on record.

Of 93 newly emerged adults held with dried alfalfa and 293 held without food, all were dead 5 weeks after emergence. Eighty-six new adults were allowed approximately

Table 2.—Alfalfa weevil adults taken in trash and soil in and near alfalfa fields during the summer months, Prince George's, Howard, and Anne Arundel Counties, Md., 1957.[a]

FIELD NO.	ALFALFA FIELD		GRAIN STUBBLE	CORN FIELDS	UNDER TREES	FENCE ROWS	WEEDY AREAS	TOTAL
	Center	Edge						
1	0	0.17	—	—	0.42	—	—	0.59
2	0	.10	—	—	2.13	—	0.06	2.29
3	0	.10	0	0	0.16	—	—	0.26
4	0	0	—	—	1.28	1.21	0	2.59
5	0	0	0	—	0.57	0.14	0.29	.91
6	0.50	0.42	—	0	—	—	—	.92
Total	0.50	0.89	0	0	4.56	1.35	0.26	7.56
Per cent of total	6.6	11.8	0	0	60.3	17.3	3.4	100.0

[a] Average numbers of adults per square foot.

3 weeks of feeding immediately after emergence but thereafter were without food until September 13, when adults were observed returning to alfalfa fields. At that time they had a survival rate of 30% as compared with 49% for 86 adults that were kept with fresh alfalfa throughout the summer. These results strengthen previous observations (Manglitz & App 1957), and are in line with studies on weevils in haystacks and baled hay by Hawley (1932), who indicated that prior feeding greatly affects weevil survival in stored hay. At any rate, it has been demonstrated that the adult weevil, after a short feeding period, can survive several months without food and apparently does so through the summer.

The adults began their return to alfalfa fields on September 11. At that time dissections showed that there had been no ovarian development during their absence from the fields, and continued observations indicated that it was not complete until late October. Therefore, the period away from alfalfa appears to be a true aestivation rather than one for the development of internal organs.

REFERENCES CITED

App, B. A. 1954. Insecticide tests to control the alfalfa weevil and the meadow spittlebug in Maryland. **Jour. Econ. Ent. 47:** 313–6.

Hamlin, J. C., F. V. Lieberman, R. W. Bunn, W. C. McDuffie, R. C. Newton, and L. J. Jones. 1949. Field studies of the alfalfa weevil and its environment. U. S. Dept. Agric. Tech. Bull. **975,** 84 pp.

Hawley, I. M. 1932. Weevils in fields, haystacks, and baled hay as possible factors in the contamination of alfalfa meal. California Dept. Agric., Spec. Publ. **115:** 94–103.

Manglitz, G. R., and B. A. App. 1957. Biology and seasonal development of the alfalfa weevil in Maryland. Jour. Econ. Ent. **50:** 810–13.

Milliron, H. E., and D. MacCreary. 1955. The alfalfa weevil in Delaware, 1953–54. Jour. Econ. Ent. **48:** 283–9.

Snow, S. J. 1928. Effect of ovulation upon the seasonal history of the alfalfa weevil. Jour. Econ. Ent. **21:** 752–61.

Sweetman, H. L., and J. Wedemeyer. 1933. Further studies of the physical ecology of the alfalfa weevil, *Hypera postica* (Gyllenhal). Ecology **14:** 46–60.

Genotypic Reactions to Boll Weevil Attack in Upland Cotton[1]

W. H. WESSLING,[2] *Genetics Faculty, North Carolina State College, Raleigh*

ABSTRACT

Mutant lines of Upland cotton carrying any two of four mutant genes, H_1, H_2, *gl* and R_1, which promised to confer a certain degree of resistance to attack by boll weevil (*Anthonomous grandis* Boh.) were tested together with lines having the same genetic background but differing by opposite alleles to the genes under test. All mutant lines showed varying degrees of resistance to boll weevil attack; the lines combining H_1 and H_2 to the highest degree, those combining *gl* and H_2, and *gl* and H_1, ranking second and third. Lines carrying the genes R_1 in combination with *gl* or H_2 were tested in a separate experiment. Both strains showed a relatively high degree of resistance. The main effects of the mutant genes, H_1, H_2 and *gl*, could only be estimated on the assumption that their interactions were unimportant. On this basis the data obtained in the experiments indicate that H_2 contributed more to resistance than H_1 while *gl* had no significant effect.

In a previous paper (Wessling 1958) an experiment was reported which showed that resistance to boll weevil attack conferred by the mutant gene, H_2, to the Upland cotton strain, Pilose, was relatively constant under different environmental conditions, *i.e.*, either in different mixtures of Pilose with a susceptible variety, All-in-One, or in the absence of susceptible plants. These results gave assurance that the degree of resistance conferred by H_2 is very little affected by the presence of susceptible plants, as long as the proportion of resistant plants in the mixed population is not too low. After this information was available further tests of resistance to boll weevil attack of various mutants and combinations of mutants were possible.

In table 1 are listed five mutant strains available for study, together with their genotypes and brief descriptions of their characteristics. It will be noted that "Pilose A" and "Pilose B" carry the same mutant, H_2. The two strains, however, have widely different genetic backgrounds, Pilose B being derived from an outcross to a strain of *Gossypium barbadense* which carried a number of factors for lint length. (The cross was made to improve lint length and lint quality of the original Pilose strain which is deficient in these properties.) The mutant, H_1, carried by MU-8 was shown to be important in conferring jassid resistance to Upland cottons in Africa (Parnell *et al.* 1949). The *gl* mutant was first described by McMichael (1954) in the Acala strains 1517 and 1-13-3 of Upland cotton. De Ridder, carrying R_1, has been used extensively for genetic and breeding purposes in this country.

PURPOSE OF EXPERIMENTS AND PROCEDURE.—Previous experiments had shown (Wannamaker 1957, Wessling 1958) or preliminary observations had suggested (Stephens 1957) that all the mutants listed in table 1 tended to reduce boll weevil damage in Upland cottons. By combining all mutants in one strain it might therefore be possible to build up a high level of resistance. As a first

[1] Contribution from the North Carolina Agricultural Experiment Station. Published with the approval of the Director as Paper No. 883 of the Journal Series. Part of a thesis presented in partial requirement for the Ph.D. degree. Accepted for publication February 24, 1958.

[2] Present address: c/o Schreiber & Söhne, 237 Boeckler Str., Braunschweig, Germany.

Table 1.—Description of mutant cotton strains used in experiments.

MUTANT STRAIN	GENOTYPE	CHARACTERISTICS
Pilose A	H_2H_2	dense pubescence over entire plant (stem, petioles, bracts, flower petals, bolls, calyx)
Pilose B	H_2H_2	characteristics like Pilose A differing in general background by genes introduced from *G. barbadense* parent
MU-8	H_1H_1	relatively long, dense hair on veins of lower leaf surface, on petioles and stem
Glandless	*glgl*	absence of oil glands in bolls, stem, hypocotyl, petioles
De Ridder	R_1R_1	presence of red anthocyanin pigment in all vegetative parts of the plant

Table 2.—Origin and genotypes of the mutant and "normal" sib lines used in Experiments I and II.

ORIGIN	LINES	
	Mutant	"Normal"
Expt. 1		
Glandless×Pilose A	$glglH_2H_2$	$GlGlh_2h_2$
Glandless×Pilose B	$glglH_2H_2$	$GlGlh_2h_2$
Glandless×MU-8	$glglH_1H_1$	$GlGlh_1h_1$
MU-8×Pilose A	$H_1H_1H_2H_2$	$h_1h_1h_2h_2$
MU-8×Pilose B	$H_1H_1H_2H_2$	$h_1h_1h_2h_2$
Expt. II		
Glandless×De Ridder	$glglR_1R_1$	$GlGlr_1r_1$
Pilose[a]×De Ridder	$H_2H_2R_1R_1$	$h_2h_2r_1r_1$

[a] Owing to the small amount of seeds available, the material from Pilose A and Pilose B was bulked in this experiment.

step in reaching this objective, the strains carrying the four mutants, H_1, H_2, *gl* and R_1, were crossed in all possible combinations. With four mutants there were six possible combinations, but since H_2 was carried both by the Pilose A and Pilose B strains, a total of nine F1 hybrids was actually made. All nine F1 hybrids were selfed and from the segregating F2 populations obtained, plants homozygous for "double mutant combinations" (H_1H_1 H_2H_2, H_2H_2 *glgl*. . . etc.) were selected. At the same time sister plants homozygous for the opposite "normal" alleles (h_1h_1 h_2h_2, h_2h_2 *GlGl*. . . etc.) in the same population were also selected. Selfed seeds from the double mutant combinations and their "normal" sibs provided the lines which were tested for resistance to boll weevil in the experiments to be described. In these experiments the "normal" sib lines were used as controls in order to exclude the possible effects of different genetic backgrounds from comparisons of the main gene effects.

EXPERIMENTAL METHODS.—It was originally planned to include a total of 18 lines (nine double mutant combinations and nine "normal" sib lines) in a single experiment. However some difficulty was experienced in obtaining sufficient seeds of the R_1 combinations in time for planting. Consequently the experiment was divided into two parts. Experiment I was confined to the five double mutant combinations not involving R_1 and their "normal" sibs. Experiment II was planted 2 weeks later and included two of the four R_1 combinations and their "normal" sibs. Seeds of the combination, H_1H_1 R_1R_1, were not obtained in time to be included in the experiment. The origin and genotypes of all these lines are listed in table 2.

The designs of both experiments were similar to that described previously (Wessling 1958) in which each plot of cotton was isolated from its neighbors by barrier plots of corn. The design, since it permits a uniform initial infestation by the weevils and prevents their subsequent movement from less resistant to more resistant plants enables small differences in resistance to be detected. Unfortunately in the experiments to be described this method of isolation failed, partly due to a poor choice of corn variety and partly to its poor early growth under abnormally hot, dry conditions. Consequently the weevils moved freely from plot to plot and some of the intended precision of the experiments was lost. Both experiments were planted in a field in the vicinity of Garner, N. C., which was made available through the courtesy of N. C. Farmer's Cooperative. Experiments I and II were planted at the middle and at the end of April, respectively. Cotton had not been grown in the field for at least 2 years and was not growing in fields closer than approximately 1 mile. Early in the season three applications of toxaphene were made until 2 weeks before the infested squares were distributed.

Infested squares were obtained and distributed in the plots. Unfortunately no rain fell during the following days and the temperature went up so high that practically all boll weevils were killed before they were able to leave the squares. Since the cotton had started flowering already, to avoid the hazard of an unequal initial infestation in different plots it was necessary either to collect adult boll weevils or to collect squares, hatch the weevils in an incubator and release them in the experiment as soon as a sufficient number was available. The latter procedure was chosen. Adult weevils were made available by Dr. R. T. Gast, after they had emerged from squares collected in North Carolina and had fed on cotton flowers for several days. One boll weevil was released for every two plants in each plot.

In both experiments a randomized block design with five blocks was used. Each block contained all lines, each plot four rows of the same line with eight plants in each row. Border rows and the end plants of each row were eliminated from scoring. The method of scoring was the same as has been described previously (Wessling 1958). The percentage of squares with egg-laying punctures was chosen as a measure of resistance because it affects the size of the future boll weevil population most. The percentages were converted to angular values (Fisher & Yates 1943) for statistical analysis.

RESULTS.—*Experiment I.*—An analysis of variance was first performed on the percentages of egg-laying punctures recorded in the whole experiment. Differences between lines were found to be highly significant. Comparisons of individual lines were then carried out as shown in table 3 where differences between transformed percentages of egg-laying punctures are analyzed by appropriate t-tests. All differences between the mutant combinations and their "normal" sib lines are shown to be significant or highly significant (comparisons 1 through 5 in

Table 3.—Comparisons of percentages of squares with egg-laying punctures in double mutant combinations and "normal" lines tested in Experiment I.

Lines	Sample Size	Mean Per Cent	Means of Angles	Differences	t-Values	P
1. Glandless×Pilose A						
glgl H_2H_2	307	53.64	47.04	18.70	8.66	<.01
GlGl h_2h_2	455	83.11	65.74			
2. Glandless×Pilose B						
glgl H_2H_2	396	52.35	46.35	19.29	9.10	<.01
GlGl h_2h_2	392	83.00	65.64			
3. Glandless×MU-8						
glgl H_1H_1	494	70.73	57.25	7.57	2.45	<.05
GlGl h_1h_1	529	81.90	64.82			
4. MU-8×Pilose A						
H_1H_1 H_2H_2	384	31.42	34.09	29.54	7.88	<.01
h_1h_1 h_2h_2	369	80.33	63.63			
5. MU-8×Pilose B						
H_1H_1 H_2H_2	335	20.76	27.10	34.23	8.05	<.01
h_1h_1 h_2h_2	426	77.00	61.33			
6. *glgl* H_2H_2	396	52.35	46.35	10.90	3.28	<.02
glgl H_1H_1	494	70.73	57.25			
7. H_1H_1 H_2H_2	384	31.42	34.09	12.26	4.38	<.01
glgl H_2H_2	396	52.35	46.35			
8. H_1H_1 H_2H_2	384	31.42	34.09	23.16	6.67	<.01
glgl H_1H_1	494	70.73	57.25			

the table). It may be concluded that all the mutant combinations tested confer a certain amount of resistance to boll weevil attack.

All differences between "normal" lines, and between lines with the same mutant gene combination but differing in genotypic background, were not significant and are not included in the table. There was therefore no indication of residual effects on resistance due to differences in genetic background, *i.e.*, most of the differences between lines was attributable to the effects of the mutant genes.

Comparisons 6, 7 and 8 in the table represent differences between lines with different mutant gene combinations. The proportion of egg-laying punctures is lowest in the lines combining the mutant genes, H_1H_1 and H_2H_2. It is significantly lower in lines carrying the mutant genes *glgl* H_2H_2 than in the line carrying the mutant genes *glgl* H_1H_1. These differences between gene combinations suggested that the individual gene components contributed to resistance to a different degree. It was decided to determine as well as possible from the data available the main effects of the individual genes. Table 4 shows the transformed mean percentages of egg-laying punctures of various genotypes. Since the experiment was not designed to test the main effects of single genes and the corresponding interactions only four out of eight mean values were available. For this reason the main effect of any one gene was confounded with interaction between the other two, as shown in table 5. In this table, the three degrees of freedom available for the four genotypes (plus confounded interactions) are partitioned, the total number of degrees of freedom (19) being obtained from the four genotypes replicated five times in the experiment.

If we may assume that the interactions confounded with the main effects are of relatively little importance, then the data indicate that the individual effects of the mutant genes, H_1 and H_2, on resistance are highly significant. An examination of the mean values in table 4, and the relative magnitudes of the mean squares in table 5

Table 4.—Transformed percentages of squares with egg-laying punctures in various mutant combinations, and mean effects of individual genes.

	Gl	*gl*	Means
H_1H_2	34.09	—	$H_1=45.67$
H_1h_2	—	57.25	$h_1=56.39$
h_1H_2	—	47.04	$H_2=40.57$
h_1h_2	65.74	—	$h_2=61.50$
Means	$Gl=49.92$	$gl=52.15$	

suggests that the gene H_2 adds more to resistance than the gene H_1 in this experiment.

It can be concluded from Experiment I that under the given conditions the mutant genes under consideration and their corresponding combinations confer different levels of resistance to boll weevil attack. The gene *gl* did not have any significant effect on boll weevil attack under these conditions. It was noticed that lines carrying the mutant gene H_2 were attacked somewhat later than the others, probably due to the fact that in these lines the hairy bracts stay tightly appressed at the margins until the growing flower bud pushes them apart. It was observed that weevils were unable to open the bracts before that stage was reached.

Boll counts in this experiment turned out to be unsatisfactory since the bolls which were set before adult boll weevils were released were removed from the plants. This was done to avoid a complete lack of development of squares under the prevailing weather conditions.

Experiment II.—The results are presented in table 6. Both mutant combinations show a significant reduction in egg-laying punctures as compared with the lines carrying the "normal" alleles. The Glandless-Red combination had significantly more egg-laying punctures than the Pilose-Red combination while the two "normal" lines were not significantly different from each other.

In this experiment the proportion of squares showing feeding punctures only was extremely low. The reduction in egg-laying punctures was therefore proportional to increase in unpunctured squares. Also it was observed that the reduction in egg-laying punctures in the Glandless-Red strain was mainly due to reduction in the early stages while the Pilose-Red strain showed a reduction in egg-laying punctures during the entire scoring period.

It is difficult if not impossible to make any reliable comparisons between Experiments I and II because of differences in planting time and environmental conditions. Since the mutant gene, *gl*, was shown in Experiment I to have no significant main effect, it seems to be

Table 5.—Analysis of variance of main effects due to genes H_1, H_2 and *gl*.

Main Effects	D.F.	Mean Squares	F	P
$H_1 \vee h_1$[a]	1	547.91	26.88	<.01
$H_2 \vee h_2$[b]	1	2190.52	102.40	<.01
$gl \vee Gl$[c]	1	25.19	1.18	>.05
Error	16	21.39		
Total	19	—		

[a] Confounded with interaction between H_2 and *gl*.
[b] Confounded with interaction between H_1 and *gl*.
[c] Confounded with interaction between H_1 and H_2.

Table 6.—Comparisons of percentages of squares with egg-laying punctures in double mutant combinations and "normal" lines tested in Experiment II.

LINES	SAMPLE SIZE	MEAN PER CENT	MEANS OF ANGLES	DIFFERENCES	t-VALUES	P
Glandless×De Ridder						
glgl R_1R_1	397	43.43	41.23	19.89	5.53	<.01
GlGl r_1r_1	460	76.68	61.12			
Pilose×De Ridder						
H_2H_2 R_1R_1	261	15.74	23.36	32.25	6.72	<.01
hh r_1r_1	434	68.10	55.61			
H_2H_2 R_1R_1	261	15.74	23.36	17.87	3.65	<.01
glgl R_1R_1	397	43.43	41.23			

fairly unlikely that it would show a large main effect under the conditions of Experiment II, though it could be of importance in interacting with the mutant gene, R_1. It seems to be safe to assume that a certain amount of resistance is conferred by the mutant gene, R_1.

DISCUSSION.—The purpose of the investigation was to test the thesis that lines of Upland cotton carrying any two of four specified mutant genes would show various degrees of resistance to boll weevil attack as compared with lines having the same genetic background but differing by opposite alleles to the mutant genes under consideration. As pointed out earlier, the original intention was to compare the lines in plots isolated from one another, but the corn plants used for this purpose, owing to poor growth, failed to provide an effective barrier. The failure of isolation probably reduced the precision of the experiments, and in fact the data obtained on differing scoring dates were found to be much more variable than had been obtained in an earlier experiment in which effective isolation had been maintained (Wessling 1958). The boll weevils were observed to migrate freely from plot to plot with consequent fluctuations in the intensity of attack. Nevertheless, highly significant degrees of resistance were demonstrated for all the combinations of mutants tested (Tables 3 and 6).

Severity of infestation.—The severity of the test was very high in both experiments. Boll weevils had already passed the maturation feeding period in the culture room and took to wing as soon as they were released in the plots. Egg-laying started immediately and because of the scarcity of squares, three or four egg-laying punctures were found frequently in the same square. Squares of susceptible lines were attacked in very early stages which accounted for an extremely severe loss of squares as soon as the experiments were started. Due to the lack of developing squares available bolls were attacked to an unusually high extent. Observations on duplicate plantings of the experimental lines which have been grown in the experimental field at North Carolina State College suggest that resistant lines might show a higher degree of resistance under heavy natural infestation than under the artificially high infestation in these experiments. It may be advisable in later experiments, when yield comparisons become of importance, to rely entirely on natural infestation. For the experiments described here extremely heavy infestation was essential.

Effect of different mutant characteristics on resistance.—Natural infestation may change the effect of resistance observed in the experiments to a certain extent. Wannamaker (1957) found that the genes, H_1 and H_2, confer resistance of equal magnitude to boll weevil attack. The writer was able on the other hand to show that these two genes differ significantly in effecting resistance. The discrepancy between these results may be explained by the fact that, in Wannamaker's experiment, boll weevils newly emerged from squares on the ground provided the initial source of infestation. In the present experiment adult weevils already capable of flight were placed directly on the plants. In the former case the young weevils may have been hampered in their efforts to reach the squares by the hairs on the surfaces of stems and petioles, while in the latter case the mechanical hindrance would presumably be less effective.

It may be, under conditions of natural infestation, that lack of glands (*glgl*) or presence of red color (R_1R_1) may affect the attraction of the boll weevil to the cotton plant and thus prove to be of more importance than appears to be the case in these experiments. Under the experimental conditions described it was not possible to test all the effects associated with nonpreference (used in the sense of Painter's definition, 1951). The boll weevils had, so to speak, no "choice" since they were put directly on the plant or close to it and did not have to search for it. It is possible, if the test had required the weevils to locate the plants, that the lines carrying the *gl* and R_1 mutants would have proved less attractive, and hence shown more apparent resistance than was observed in these experiments.

The effect of red plant color might not be due entirely to a smaller degree of attraction but might have also an antibiotic effect. It is somewhat difficult to evaluate Experiment II with respect to the effectiveness of the gene, R_1, conferring red plant color. The late planting and drought during the early growing season retarded the development of the plants in such a way that very little of the soil surface was covered and protected from direct sunlight. This increased the temperature within the plots, accelerated movement of the boll weevils (Fenton & Dunnam 1929), and caused natural shedding of squares. Comparisons between Experiment I and II are therefore invalid. In spite of these difficulties the conclusions drawn from the results obtained from Experiment II are believed to be justified since the mutant combinations and "normal" lines were strictly comparable with regard to environmental conditions.

Nature of resistance to boll weevil attack.—It was not the purpose of this investigation to determine the nature of resistance to boll weevil attack in resistant Upland cotton strains. For this reason only little information can be supplied from but a few personal observations. It is not believed that tolerance is exerted by any of the mutant lines under consideration. Most probably the main cause of resistance is nonpreference, while antibiosis may or may not play a part in causing resistance. The writer noticed that boll weevils flew to a plant having the characteristics conferred by the mutant genes, H_1H_1 H_2H_2. In some cases the weevils left the plants after a short search for squares, in others they stayed on the plant and kept moving until they finally located a suitable square. If the square was still small with the bracts tightly closed efforts of the weevil to get to the flower bud were usually unsuccessful. After a period of fruitless efforts weevils attempted to puncture through the bracts, to puncture

leaves, and petioles. In general the main effect of hairiness was to distract the boll weevils or to make their attempts to locate and puncture squares more difficult. Since neither of the other two possible causes of resistance can be applied to these observations nonpreference can be assumed as the major cause of resistance.

In the case of characteristics conferred by the R_1 and $g1$ genes the writer has to rely on observations of other investigators. Hunter & Pierce (1912) make reference to a study by Jones (1907) who investigated the attraction of boll weevils by different colors and found that red light was approximately half as attractive to the boll weevil as blue and green light. This might be taken as a suggestion that resistance due to red plant color is also caused mainly by nonpreference.

Stanford & Viehover (1918) suggested that volatile oil excreted by the interior glands of the cotton plant was the cause of attraction of the boll weevil to the cotton plant. This indicates that, in the case that the mutant gene, *gl*, should confer resistance under natural infestation, nonpreference might be the main cause of it.

Since the writer's observations are very few, and observations of other authors are not very conclusive the only safe statement which can be made is that we do not know what the actual causes of resistance to boll weevil attack are in the mutant lines of Upland cotton under consideration until more detailed investigations may supply a satisfactory answer.

Based on the results of the experiments considered here investigations of resistance to boll weevil attack may be carried on along the following lines: 1. Investigations of the causes of resistance; 2. combination of all genes conferring various degrees of resistance to boll weevil attack in Upland cotton in a single strain; 3. improvement of fiber properties and yield of the resistant strains.

References Cited

Fenton, F. A., and E. W. Dunnam. 1929. Biology of the cotton boll weevil at Florence, S. C. U. S. Dept. Agric. Tech. Bull. 112.

Fisher, R. A., and F. Yates. 1943. Statistical tables for biological, agricultural and medical research. London, Oliver and Boyd. 98 pp.

Hunter, W. D., and W. D. Pierce. 1912. The Mexican cotton boll weevil. U. S. Dept. Agric. Ent. Bull. 114.

McMichael, S. C. 1954. Glandless boll in Upland cotton and its use in the study of natural crossing. Agron. Jour. 46(11): 527–9.

Painter, R. H. 1951. Insect Resistance in Crop Plants. New York, Macmillan Co., 520 pp.

Parnell, F. R., H. E. King, and D. F. Ruston. 1949. Jassid resistance and hairiness of the cotton plant. Bull. Ent. Res. 39: 539–75.

Stanford, E. E., and A. Viehover. 1918. Chemistry and histology of the glands of the cotton plant, with notes on the occurrence of similar glands in related plants. Jour. Agric. Res. 13: 419–35.

Stephens, S. G. 1957. Sources of resistance of cotton strains to the boll weevil and their possible utilization. Jour. Econ. Ent. 50(4): 415–8.

Wannamaker, W. K. 1957. The effect of plant hairiness of cotton strains on boll weevil attack. Jour. Econ. Ent. 50(4): 418–23.

Wessling, W. H. 1958. Resistance to boll weevil in mixed populations of resistant and susceptible cotton plants. Jour. Econ. Ent. 51(4): 502–6.

Aster-Yellows Virus Infection and Transmission by *Macrosteles fascifrons* (Stål) with Reference to Lettuce-Yellows Prevention[1]

R. G. Strong[2] and W. A. Rawlins, *Cornell University, Ithaca, New York*

Abstract

Transmission studies with the eastern strain of the aster-yellows virus, *Chlorogenus callistephi vulgaris* Holmes, showed that after 18 days' incubation of the virus in six-spotted leafhoppers, *Macrosteles fascifrons* (Stål), no differences were found in infectivity of viruliferous leafhoppers with infection feeding periods of 3, 6, and 10 days on China aster, *Callistephus chinensis*. With 18 days' virus incubation, two viruliferous insects per plant were as successful as larger numbers in inoculation of all healthy plants. Infective insects resting on plants for 10 minutes transmitted virus almost as readily as those caged on plants for 6 hours, but the percentage of infection was considerably higher for insects caged on plants for 24 hours. Most infective leafhoppers inoculated only one plant per 24-hour period of individual confinement to groups of plants. After becoming infective, 70 to 78% of viruliferous leafhoppers inoculated all plants on which they were confined individually for 12 to 48 hours in serial transfers. Such consistent infection was not obtained with 1-hour confinement periods. Five commonly grown varieties of lettuce and one variety of China aster were equally susceptible to aster-yellows virus infection under experimental conditions existing.

A high potential yellows incidence is considered essential to make good comparisons between insecticides in their ability to prevent transmission of aster-yellows virus, *Chlorogenus callistephi vulgaris* Holmes, to lettuce by six spotted leafhoppers, *Macrosteles fascifrons* (Stål). The average incidence of lettuce yellows in most fields in New York State is usually low, however, presumably due to a very small proportion of infective leafhoppers in the natural population.

The problem of obtaining a consistently high potential yellows incidence in field experiments was solved by using controlled populations of viruliferous leafhoppers with a high percentage of infective individuals. Large numbers of viruliferous leafhoppers were required. Leafhoppers could be mass-produced without difficulty,

[1] Accepted for publication February 24, 1958.

[2] Present address: Department of Entomology, University of California, Citrus Experiment Station, Riverside, Calif.

but information on virus infection and transmission supplementing the work of Black (1941), Kunkel (1924, 1926, 1954), Maramorosch (1952, 1953) and others, was needed before a high percentage of infective leafhoppers could be assured in cultures. Furthermore, some additional facts were necessary before results from field experiments with insecticides could be properly interpreted. The nature of the problem required such heterogenous studies as those reported in this paper.

TERMINOLOGY.—Terminology varies considerably in reports dealing with virus transmission and often leads to confusion. In an attempt to be consistent, the authors have used terms as defined by Maramorosch (1950) in describing the relationship between the plant, vector, and virus.

GENERAL EXPERIMENTAL PROCEDURES.—The eastern strain of the aster-yellows virus originally came from diseased lettuce plants (*Lactuca sativa*) through six-spotted leafhoppers to periwinkle plants (*Vinca rosea*) which were kept in a greenhouse for a permanent source of the virus during the investigations. The virus was transmitted to China aster plants (*Callistephus chinensis*) as needed, and aster plants were used as source plants for infection feeding of leafhoppers.

Six-spotted leafhoppers were cultured and handled as described by Strong & Rawlins (1957). Non-viruliferous, unsexed adult leafhoppers ranging from 1 to 10 days in age, were placed on virus source plants for the desired period of infection feeding. They were then removed and held on barley, a nonsusceptible plant, until ready for use. The incubation period for the virus in leafhoppers was considered as the time from initial infection feeding to confinement on test plants.

Circular cellulose acetate cages, $3\frac{3}{4}$ inches wide and 5 inches high, were used to confine leafhoppers to plants for test feeding. The tops of the cages and the two air vents, each 1 inch in diameter, on opposite sides of the cages were covered with 24-×34-mesh nylon tulle. This permitted ample circulation of air and prevented moisture accumulation inside the cages. A transfer opening for introducing insects was closed with a cork stopper when not in use. After test feeding by the leafhoppers, the test plants were held for about 30 days before recording the yellows incidence. All records of virus transmission were based on the yellows incidence in plants alive at the end of an experiment. A few plants died during the holding period but none of these showed symptoms of aster yellows.

Specific details in regard to procedures for each experiment are given later. Unless otherwise stated, all tests were conducted in a greenhouse with a temperature range of 70° to 90° F.

MINIMUM INFECTION FEEDING TIME REQUIRED ON CHINA ASTER SOURCE PLANTS FOR MAXIMUM PERCENTAGE OF INFECTIVE INSECTS.—The most suitable infection-feeding time required by leafhoppers on diseased China aster was determined by comparing the effect of varying lengths of infection feeding on (1) the ability of individual leafhoppers to transmit aster-yellows virus, and (2) the incubation period of the virus in the insect. Leafhoppers of the same age and culture background were used.

Non-viruliferous leafhoppers were placed on three diseased China aster plants. Each plant was infested with 100 leafhoppers. The insects were allowed to feed on the infected plants for 1, 3, 6, and 10 days, respectively. After the allotted infection feeding period for the leafhoppers concerned, 25 leafhoppers were taken from each source plant. These were held on barley until ready for test feeding.

Ten days from the time of initial infection feeding on China aster source plants, individual leafhoppers were caged over healthy young China aster test plants and left for 24 hours. Part of the insects feeding for 10 days on infected plants were moved directly to test plants, while the remaining insects were held on barley. At the end of 24 hours' test feeding all leafhoppers were removed from test plants and again held on barley. Thirty plants were used for each sample of 30 insects from each of the infection feeding periods involved in the experiment. Ten unexposed plants were left for checks. The procedure was repeated at the end of 14, 18, and 22 days from the time of initial infection feeding.

The percentages of test plants infected by insects from each infection-feeding period on China aster source plants are given in table 1. The proportion of transmitters at the end of 22 days' incubation of the virus in the insect seemed to be correlated with the length of infection feeding periods. The longer the insects were confined on the virus source plants, the more insects that became infective. However, at the end of 18 days there were little or no differences as a result of 3, 6, or 10 days of infection feeding. The incubation period appeared to be slightly shorter in insects feeding for more than 1 day on infected plants.

Six-spotted leafhoppers readily fed and bred on aster plants, and viruliferous insects were quite vigorous during the test periods. Very little mortality occurred, although some was expected because of the frequent handling of the insects.

MINIMUM NUMBER OF VIRULIFEROUS LEAFHOPPERS AND LEAST INCUBATION PERIOD OF VIRUS NECESSARY FOR 100% INOCULATION.—The necessity for a high potential inoculation of test plants in experiments to prevent aster-yellows virus transmission by controlling leafhoppers has been explained. An experiment was conducted to determine the minimum number of viruliferous leafhoppers necessary for 100% inoculation of healthy plants in relation to the length of the incubation period of the virus in the insects.

Non-viruliferous six-spotted leafhopper adults were

Table 1.—Effect of infection feeding period on China aster source plants and of succeeding incubation period on the ability of the six-spotted leafhopper to transmit aster-yellows virus to healthy China aster.

INFECTION FEEDING PERIOD ON DISEASED PLANTS (DAYS)	PERCENTAGE OF HEALTHY CHINA ASTER PLANTS INFECTED AFTER VIRUS INCUBATION PERIOD (DAYS) IN INSECT			
	10	14	18	22
1	20.0	30.0	43.3	70.0
3	16.7	21.4	73.3	76.7
6	21.4	20.0	73.3	86.7
10	18.5	36.7	70.0	90.0

Table 2.—Effect of length of virus incubation period in the six-spotted leafhopper on percentage of China aster plants infected in two tests with varying numbers of viruliferous leafhoppers.[a]

Insects per Plant	Percentage of Healthy China Aster Plants Infected After Virus Incubation Period (Days) in Insect						
	8	10	14	16	18	20	22
			Test No. 1				
1	2/50	2/50	11/50				67/50
2	5/40	13/40	20/40				100/40
3	10/30	4/30	26/28				100/25
4	15/20	10/20	26/20				100/13
5	10/10	25/10	20/10				100/10
			Test No. 2				
1			57/50	53/30	87/30	77/30	90/30
2			39/40	60/25	100/25	96/25	100/25
3			67/10	90/10	100/10	90/10	100/10
4			43/10	90/10	100/10	100/10	100/10
5			80/10	100/10	100/10	100/10	100/10

[a] Data presented as percentage of plants infected/number of test plants.

confined to diseased China aster plants for 3 days. After the 3-day infection feeding period, leafhoppers were moved to barley and held until ready for test feeding. At the end of 8 days' incubation of the virus in the insects, the viruliferous leafhoppers were taken from barley and caged over healthy young China aster test plants. The insects were removed from test plants after 24 hours of test feeding. This procedure was followed with other insects at the end of 10, 14, and 22 days of virus incubation. The number of insects used per plant, the number of test plants employed, and the percentage of test plants infected are given under Test No. 1 in table 2. A great change in the infective ability of viruliferous leafhoppers is shown between 14 and 22 days' incubation period of the virus in the vector. This point was of considerable importance.

An additional test was conducted (Test No. 2, table 2) using the same procedure as that already described, except that test feedings were made at the end of 14, 16, 18, 20, and 22 days of virus incubation in the insects. The data for individual insects in table 2 for the incubation periods common to both groups of insects used, namely, 14 days and 22 days, leave no doubt that the two test groups of insects differed considerably in their infective ability and in the virus incubation period. This was not considered unusual, however, as Kunkel (1926) also obtained incubation periods which varied somewhat in different experiments with different groups of insects.

With 22 days' virus incubation in Tests 1 and 2, two leafhoppers transmitted the virus with as much certainty as it was transmitted by groups of 3, 4, or 5. In Test No. 2, pairs of leafhoppers were as successful as the larger numbers in inoculating 100% of the plants by the end of 18 days' incubation.

Length of Plant Exposure to Individual Viruliferous Leafhoppers and Frequency of Infection.—When a viruliferous leafhopper rests on a plant it may or may not feed. If the plant should be covered with an insecticide, the leafhopper may be killed before it has a chance to feed and transmit the virus. A test was made to determine the relation between the time viruliferous leafhoppers are in contact with plants and the frequency of aster-yellows virus infection.

Twenty-five viruliferous leafhoppers were taken from each of eight cultures and "pooled" on fresh barley. The infection feeding period was not less than 3 days, and the incubation period of the virus in the insects was not less than 20 days. Individual insects were taken from the group "pooled" over barley, and single healthy China aster test plants were exposed to these individual insects for varying periods. The exposure period was measured from the time the leafhopper first rested on the plant until it was removed, whether it remained on the plant for the entire time or not.

Table 3.—Effect of period of exposure of China aster plants to individual viruliferous six-spotted leafhoppers on percentage of infection by aster-yellows virus.

Exposure Period of Plants to Viruliferous Leafhoppers	No. of Plants Exposed	Percentage of Plants Infected
10 minutes	50	24.0
30 minutes	50	8.0
1 hour	50	24.0
3 hours	50	24.0
6 hours	50	32.0
24 hours	30	66.7

The exposure periods, the total number of test plants exposed to individual insects, and the percentages of plants infected are given in table 3. The data show that viruliferous leafhoppers resting on plants for 10 minutes transmitted virus almost as readily as those in contact with plants for periods as long as 6 hours. Virus transmission in the 24-hour group was considerably higher. These data indicate that an insecticide must kill viruliferous leafhoppers quickly to prevent infection of plants.

Plants Inoculated Per Insect in 24 Hours.—An attempt was made to determine the number of plants one infective leafhopper would inoculate in a 24-hour period. Thirty insects were taken from the group "pooled" on barley for the preceding experiment and were caged individually over 5-plant groups of healthy young China aster test plants for 24 hours. Thirty 8-inch flower pots, each containing five China aster test plants, were used for exposure to individual viruliferous insects. Five additional unexposed groups of plants were used for checks.

Sixteen of the 30 viruliferous leafhoppers released on aster plants transmitted aster-yellows virus during the 24-hour period of confinement. Of these only one leafhopper infected two plants, and 15 inoculated one plant per infective insect. Although insects were caged on plants held in the greenhouse, the results obtained indicate that six-spotted leafhoppers do not feed extensively on different plants in any one 24-hour period.

Consistency of Plant Inoculations by Individual Leafhoppers.—This experiment was conducted to determine how consistently individual infective six-spotted leafhoppers inoculate plants in a series of exposures. It was divided into two parts.

Plants Exposed to Viruliferous Insects for Periods of 12 to 48 Hours.—Insects, taken from five cages of viruliferous leafhoppers, had a minimum infection feeding period of 3 days. The minimum incubation period of the virus in the insects was 18 days. Leafhoppers were caged individually for a period of 12 to 48 hours on healthy young lettuce test plants. Then each leafhopper was moved to another healthy lettuce plant for a 12- to 48-hour test feeding period. This procedure was repeated until at least five plants had been exposed, in succession, to

each leafhopper. Not more than 10 plants were exposed to any leafhopper.

After test feeding by the leafhoppers, lettuce plants were held outdoors a short distance from the insectary where tests were conducted. Plants were frequently sprayed with malathion insecticide to reduce possible infection from other sources during the holding period.

The infection records for individual leafhoppers have been summarized in table 4. Viruliferous leafhoppers tested on a series of plants are shown in decreasing numbers from five plants to 10 plants per series. This is because some leafhoppers died after five or more serial transfers to lettuce plants, others escaped, and others were started too late to use on 10 plants in a series in this test. Yellows incidence records were made only for test-plant series where leafhoppers survived five or more transfers. This occurred with 45 out of 50 insects used. Thus, in considering these data, direct comparisons should not be made between number of plants in a series. However, the similarity of results regardless of the number of plants in a series is considered worthy of notice.

Of the 45 viruliferous leafhoppers tested, 43 transmitted virus to one or more plants. Some did not transmit the virus to the first plants of a series. In table 4 note that only a small percentage of infective leafhoppers inoculated all plants of the group. A much higher percentage of infective leafhoppers inoculated all plants after the first infection.

Plants Exposed to Viruliferous Insects for 1-Hour Periods.—Thirty viruliferous leafhoppers with a minimum infection feeding period of three days and a minimum incubation period of 20 days for the virus were caged individually for 1 hour on healthy young China aster test plants. These insects were transferred serially at hourly intervals to fresh test plants until each insect had been transferred to 10 plants. Seventeen of the 30 viruliferous insects inoculated one or more test plants. Of these 17 infective insects, 2 inoculated only 1 plant per insect, 4 inoculated 2 plants, 4 inoculated 3 plants, 4 inoculated 4 plants, 1 inoculated 5 plants, and 2 inoculated 6 plants. Nine of the infective insects transmitted to 2 plants in consecutive order of exposure, but only 3 infective insects transmitted to 3 plants, only 1 to 4 plants, and only 1 to 5 plants in consecutive order of exposure. A comparison of these data with those presented in the preceding test shows that consistent infection of plants depends on the length of time each plant is exposed to an infective leafhopper.

DIFFERENCES IN YELLOWS SUSCEPTIBILITY OF LETTUCE AND CHINA ASTER.—China aster was more appropriate than lettuce for greenhouse work, so this plant was used for most transmission experiments. In addition, basic work reported in the literature on the aster-yellows virus in relation to its plant host is concerned primarily with China aster. The present investigations, however, were for the purpose of developing techniques for leafhopper control and the prevention of aster yellows in lettuce.

Before transposing information concerning asters to lettuce, it was considered important to learn if there were significant differences between China aster and the commonly grown varieties of lettuce in susceptibility to aster-yellows virus. An experiment was conducted to determine differences, if any, between a variety of China aster (Early White Giant) and five commonly grown varieties of lettuce in New York (Great Lakes 659, Cos or Romaine, Cornell 456, Pennlake, and White Boston).

Table 4.—Transmission of aster-yellows virus to series of lettuce plants by individual six-spotted leafhoppers after test feeding periods of 12 to 48 hours.

	Plants of Series Consecutively Exposed to Individual Viruliferous Insects					
	5	6	7	8	9	10
Leafhoppers tested	45	44	41	34	33	31
Percentage infective	86.6	88.6	87.8	88.2	93.9	96.8
Percentage infective insects inoculating all plants in series	30.8	28.2	27.8	30.0	26.7	20.0
Percentage infective insects inoculating all plants after first infection	71.8	74.4	77.8	76.7	74.2	70.0

Healthy young lettuce and China aster plants of the same age were used for test plants. The experiment was designed as a randomized complete block with three replicates, each replicate containing 10 plants of each variety. Thirty unexposed China aster plants were held as checks. Two viruliferous leafhoppers were caged over each plant for 24 hours. Test plants were held in the greenhouse for 20 days before making yellows incidence records.

All test plants exposed to viruliferous leafhoppers became infected. Thus, lettuce and aster plants were equally susceptible to aster-yellows virus infection under the conditions of the experiment. Furthermore, there were no differences in the time for symptom development in test plants held in the greenhouse. It was concluded that basic knowledge on the aster-yellows virus is applicable to lettuce although China aster may have been used as the test plant.

REFERENCES CITED

Black, L. M. 1941. Further evidence for multiplication of the aster-yellows virus in the aster leafhopper. Phytopath **31**: 120–35.

Kunkel, L. O. 1924. Insect transmission of aster yellows. (Abstract) Phytopath **14**: 54.

Kunkel, L. O. 1926. Studies on aster yellows. Amer. Jour. Bot. **13**: 646–705.

Kunkel, L. O. 1954. Maintenance of yellows-type viruses in plant and insect reservoirs. *In* Hartmen, Frank W., *et al.*, eds., *The Dynamics of Virus and Rickettsial Infections.* (pp. 150–63) Blakiston, N. Y.

Maramorosch, Karl. 1950. Influence of temperature on incubation and transmission of the wound-tumor virus. Phytopath **40**: 1071–93.

Maramorosch, Karl. 1952. Multiplication of aster-yellows virus in its vector. Nature **169**: 194–5.

Maramorosch, Karl. 1953. Incubation period of aster-yellows virus. Amer. Jour. Bot. **40**: 797–809.

Strong, R. G., and W. A. Rawlins. 1957. Mass rearing of six-spotted leafhoppers. Jour. Econ. Ent. **50**(1): 76–79.

Insecticidal Residues in Milling Fractions from Wheat Treated with Methoxychlor, Malathion, and Lindane[1]

JOHN H. SCHESSER, W. E. PRIDDLE,[2] and E. P. FARRELL[3]

ABSTRACT

Milling tests were conducted to determine the insecticide residue in flour derived from wheat to which three candidate protective treatments had been applied as dusts or sprays at several rates. Some lots were milled 9 to 10 days after treatment and others after aging 18 to 24 months. Samples analyzed were the wheat just before and after cleaning, and the flour, bran, shorts, and germ. In most cases, cleaning removed only a small amount of insecticide. The highest residues were in the bran and shorts. Very little insecticide carried over into the flour, 0 to less than 0.3 p.p.m. from malathion applied at 2.5 to 7.5 p.p.m.; 0.3 to 1.8 p.p.m. from methoxychlor applied at 5 to 50 p.p.m.; and 1.3 to 2.6 p.p.m. from lindane applied at 2.5 to 7.5 p.p.m.

With the accumulation of large reserve stocks of grain during the past few years, handling and storage practices have changed. Grain is stored for much longer periods and as a result improved methods of insect control are required. The emphasis is now on preventive rather than curative methods in protecting stored grain from insect attack.

In line with this trend, protective treatments of stored wheat have been investigated since 1952. As a result of these investigations, formulations of synergized pyrethrins are recommended for application as dusts and sprays to stored wheat to protect it from insect attack. There are a number of other insecticides that are more stable and longer lasting than pyrethrum. Some of these other insecticides have a relatively low level of toxicity to warm-blooded animals, and it is possible that they can be used safely on wheat and that residue tolerances can be established to permit their use. Preliminary tests with some of these materials have shown promise of effectiveness from the standpoint of killing established infestations as well as preventing new ones.

A necessary part of the development of protective treatment for stored wheat is the determination of the fate of the insecticidal residue on the wheat when it is milled. This paper reports the results of milling tests of wheat treated with methoxychlor, malathion, and lindane. The treatment studies were conducted by the Stored-Grain Insects Laboratory, Manhattan, Kansas. The analysis of residue was done by the Chemical Unit of the Stored-Product Insects Section at Savannah, Georgia. The wheat was milled in the pilot mill of Kansas State College at Manhattan.

MATERIALS AND METHODS.—The wheat used in the test was furnished by Commodity Credit Corporation from reserve stocks stored in circular metal bins, each having a capacity of 3,250 bushels. Each lot of wheat consisted of 30 bushels and was bagged as it ran from the storage bin in clean, 10½-ounce, lintless jute bags for transporting to the mill. Precautions were taken to protect each lot of wheat from contamination by other insecticides.

Samples for residue analyses were drawn from the last two-thirds of each 30-bushel lot as it was being milled. Samples of the whole grain before and after cleaning and tempering were taken directly from an auger inside the mill. Samples of the milling fractions were taken at points in the mill where the desired sample could be easily obtained.

Each sample was composed of a blended composite of 25 sub-samples taken at 5-minute intervals during the last two-thirds of each 30-bushel run. Each composite sample amounted to about 1 gallon, with the exception of the germ sample. Since the total amount of germ for each 30-bushel lot amounted to less than 1 gallon, the entire amount from the last two-thirds of the run was collected and used in the same manner as the composite samples of the other milling fractions.

In addition to the germ sample, samples were obtained from the bran, shorts, and flour fractions. The flour was a composite of all flour streams rather than separate samples of each flour stream.

Each day's grinding was started by milling a 30-bushel untreated control lot of wheat. This amount was necessary to "warm-up" the mill. "Warm-up" refers to making all the necessary settings and adjustments in the mill. Samples were taken from the untreated control lots and submitted in the same manner as those from the treated lots of wheat.

Each composite sample was divided with a Boerner Sample Divider. Two quarts were sealed in a plastic bag and shipped to Savannah for residue analyses. The remainder of the sample was sealed in a clean glass jar and held at the Manhattan laboratory as a reserve until the results of analyses were received.

The chemical analyses of the insecticidal residues were made by the methods that appear to give the greatest accuracy in the presence of grain products. The malathion was determined by the method of Norris *et al.* (1954) as modified in a private communication (1955) for use with milled wheat products: wheat, flour, bran, shorts, etc. The methoxychlor was determined by the colorimetric method of Claborn & Beckman (1952), and the lindane by that of Schechter & Hornstein (1952).

The wheat used in this test was from two dates of treatments: (1) wheat treated 18 to 24 months previously and designated as "aged" treatments, and (2) wheat treated a few days before milling and designated as "fresh" treatments.

Several levels of application rates were used in the tests in order to observe whether the residues followed in the same comparative order as the application rates.

The malathion spray formulations were made from a

[1] Contribution No. 297, Dept. of Flour and Feed Milling Industries, Kansas Agricultural Experiment Station. Accepted for publication February 25, 1958.

[2] Stored-Product Insects Section, Agricultural Marketing Service, U. S. Department of Agriculture.

[3] Department of Flour and Feed Milling Industries, Kansas State College, Manhattan.

57% emulsifiable concentrate and the lindane sprays from a 20% emulsifiable concentrate. These were applied at approximately 5 gallons of dilute spray to 1,000 bushels of wheat. A refined grade of malathion was used.

The dusts were all made with wheat flour as the carrier at concentrations of 0.5 and 1% of malathion, 1, 2, 3, and 5% of methoxychlor, and 0.5% of lindane. These dusts were applied at rates of 30, 60, and 90 pounds per 1,000 bushels of wheat as necessary to obtain the desired dosage of toxicant in parts per million.

Malathion

1. Malathion (57% emulsifiable concentrate) ..... 6 grams
   Water ..... 744 ml.
   Application rate: above amount applied to 30 bushels of wheat
2. Malathion 0.5%
   Wheat flour 99.5%
   Application rate: 30, 60, and 90 lb./1,000 bu. for dosages of 2.5, 5, and 7.5 p.p.m. of malathion

Lindane

1. Lindane (20% emulsifiable concentrate) ..... 8.5 grams
   Water ..... 741.5 ml.
   Application rate: above amount and 3 times above amount applied to 30 bushels for dosages of 2.5 and 7.5 p.p.m. of lindane

AGED TREATMENTS

Malathion

1. Malathion (57% emulsifiable concentrate) 9 liquid ounces
   Water ..... 5 gallons
   Application rate: 5 gal./1,000 bu.
2. Malathion 0.5%
   Wheat flour 99.5%
   Application rate: 30 lb./1,000 bu. for dosage of 2.5 p.p.m. of malathion
3. Malathion 1.0%
   Wheat flour 99.0%
   Application rate: 30 lb./1,000 bu. for dosage of 5 p.p.m. of malathion

Methoxychlor

1. Methoxychlor 1.0%
   Wheat flour 99.0%
   Application rate: 30 and 60 lb./1,000 bu. for dosages of 5 and 10 p.p.m. of methoxychlor
2. Methoxychlor 2.0%
   Wheat flour 98.0%
   Application rate: 30 lb./1,000 bu. for dosage of 10 p.p.m. of methoxychlor
3. Methoxychlor 3.0%
   Wheat flour 97.0%
   Application rate: 30 lb./1,000 bu. for dosage of 15 p.p.m. of methoxychlor
4. Methoxychlor 5.0%
   Wheat flour 95.0%
   Application rate: 30 and 60 lb./1,000 bu. for dosages of 25 and 50 p.p.m. of methoxychlor

Lindane

1. Lindane 0.5%
   Wheat flour 99.5%
   Application rate: 60 lb./1,000 bu. for dosage of 5 p.p.m. of lindane

RESULTS.—The amounts recovered are given in table 1. In two-thirds of the cases, the amount of insecticide recovered from the treated whole wheat was one-half or less of the amount of insecticide applied.

The results from analyzing the milling fractions for insecticidal residues are also given in table 1. From these figures it is apparent that most of the retained insecticide

Table 1.—Insecticidal residue found during milling and in the milling fractions of wheat treated with malathion, methoxychlor, and lindane.

Lot No.	Formulation	Dosage (p.p.m.)	Date		Recovery of Insecticide (p.p.m.) from:					
					Whole Grain		Milling Fractions			
			Treated	Milled	Before Cleaning	After Cleaning	Shorts	Bran	Germ	Flour
					Aged Malathion					
1	Dust	2.5	9/ 9/55	4/25/57	<0.2	<0.2	2.8	2.0	0.8	<0.2
2	Dust	5.0	9/ 2/55	5/22/57	.4	.4	.8	< .2	< .2	< .2
3	Spray	5.0	10/10/55	5/22/57	.4	< .2	< .2	.4	< .2	.0
					Fresh Malathion					
4	Dust	2.5	6/ 4/57	6/13/57	0	0	1.0	1.2	.5	0
5	Dust	5.0	6/ 4/57	6/13/57	1.2	0	1.3	2.4	1.2	0
6	Spray	5.0	6/ 3/57	6/13/57	1.2	0	3.0	3.4	1.3	0
7	Dust	7.5	6/ 4/57	6/13/57	3.2	.8	2.1	2.1	1.5	< .3
					Aged Methoxychlor					
8	Dust	5.0	9/20/55	1/29/57	2.7	1.8	3.0	6.9	3.9	.6
9	Dust	10.0	10/ 1/55	5/22/57	5.0	5.4	7.8	14.9	7.2	.9
10	Dust	10.0	7/15/54	4/25/57	7.5	5.9	12.3	19.8	14.9	1.5
11	Dust	15.0	10/18/55	5/22/57	7.0	5.9	10.1	17.7	8.7	1.5
12	Dust	25.0	7/ 5/55	1/29/57	14.1	10.8	15.6	27.3	18.8	1.8
13	Dust	25.0	7/16/54	4/25/57	10.8	8.7	20.0	33.0	22.1	.6
14	Dust	50.0	7/17/54	4/25/57	33.6	27.0	43.2	88.7	14.1	.3
					Aged Lindane					
15	Dust	5.0	6/17/53	1/29/57	8.6	7.8	13.0	49.5	11.8	2.3
					Fresh Lindane					
16	Spray	2.5	6/ 3/57	6/13/57	3.5	3.0	7.9	8.0	9.4	2.6
17	Spray	7.5	6/ 4/57	6/13/57		4.0	8.4	16.0	15.0	1.3
					Untreated Controls					
18	—	—	—	4/25/57	0	0	0	0	0	0
19	—	—	—	1/29/57	0	0	0	0	0	0
20	—	—	—	6/13/57	0	0	0	0	0	0

was in the pericarp and was recovered from the bran and shorts.

Malathion was recovered in amounts far below the applied dosage. The residues were found in the shorts and bran, and less than 1 p.p.m. was recovered in the flour fraction.

The results obtained from methoxychlor treatments followed about the same pattern as the malathion tests. The residues in flour were all below 2 p.p.m.

In the tests with fresh lindane, the residue pattern was nearly the same as for the other fresh treatments, except that greater residues were found in the flour. One other exception noted in the lindane tests was that the aged treatment did not follow the same pattern as malathion and methoxychlor in the recovery of insecticidal residue from the whole grain. It had a concentration of 8.6 p.p.m. as compared with 5 p.p.m. applied. Similar results were obtained in a previous milling test. Possible explanations for this are that either the insecticide was not uniformly distributed when applied, or that it was translocated in the grain mass during the storage period by unequal heating, convection currents, or other unknown factors.

ANALYSES OF RESULTS:

1. About one-half or less of the applied malathion or methoxychlor was recovered in the sample taken just prior to the cleaning process. The lindane samples showed residues greater than the application rates.
2. With the exception of fresh malathion treatments, the cleaning process removed only a small amount of insecticides, an indication that the insecticides must have been in the grain.
3. The largest portion of the residual insecticides appeared in the shorts and bran, an indication that the residues were retained in the pericarp.
4. Very small amounts of the insecticides were carried through the milling process into the flour.

REFERENCES CITED

American Cyanamid Company. 1955. Private communication.

Claborn, H. V., and H. F. Beckman. 1952. Determination of 1, 1, 1-trichloro 2, 2-bis (p-methoxyphenyl) ethane in milk and fatty materials. Analyt. Chem. **24:** 220–22.

Norris, M. V., W. A. Vail, and P. R. Averell. 1954. Pesticide residues. Colorimetric estimation of malathion residues. Jour. Agric. and Food Chem. **2:** 570–73. 1955.

Schechter, M. S., and J. Hornstein. 1952. Colorimetric determination of benzene hexachloride. Analyt. Chem. **24:** 544.

Comparative Responses of Five Species of Spider Mites to Four Acaricides[1]

MOHAMMED RAMADAN ABO-EL-GHAR[2] and H. BRUCE BOUDREAUX[3]

ABSTRACT

Sulphur, Aramite, parathion and Phostex were tested for toxicity to various stages of five species of spider mites: *Tetranychus tumidus* Banks, *T. gloveri* Banks, *T. desertorum* Banks, *T. cinnabarinus* Boisduval and *T. lobosus* Boudreaux. Mites used were from non-resistant stocks. Parathion was the most toxic compound for all stages, sulphur the least. Aramite and Phostex were approximately equal in toxicity. The most susceptible species was *T. desertorum;* the least was *T. tumidus*. The three other species were approximately equal in susceptibility. Based upon acaricidal responses of the various stages, *T. tumidus* and *T. gloveri* were very similar, while *T. lobosus* and *T. cinnabarinus* formed another group, with *T. desertorum* falling in an isolated position. This agrees with the taxonomy of the species based upon morphology. Parathion and Phostex were similar in the responses of adults compared with nymphs. Sulphur and Aramite were different from each other and from the other two compounds in the responses of adults compared with nymphs. Eggs were always the most resistant stage. In evaluating acaricides, all stages of the life cycle must be taken into consideration.

It is generally recognized that a pest species causing damage should be determined before remedies are recommended. Failure to recognize the importance of an exact determination is a frequent cause of failure in control. This applies to spider mites as well as to insects.

In their work with *Septanychus texazona* McG. = (*T. desertorum* Banks) and *T. bimaculatus*, Mistric & Rainwater (1952) showed that adults of *S. texazona* were significantly more susceptible to all sulphur and phosphorus compounds tested than adults of *T. bimaculatus*. Gaines *et al.* (1950) have shown that sulphur compounds (Aramite and sulphur dusts) proved more toxic to *Septanychus texazona* McG. than to *T. bimaculatus* Harvey while organic phosphates were nearly equal in effectiveness against both species. It seems possible that their *T. bimaculatus* was misidentified, and that they were perhaps testing another species, since at that time all two-spotted mites in this country were called *T. bimaculatus* (see Boudreaux 1956). It has been found by Melvin & Earle (1948) that a species of *Paratetranychus* was approximately five times as resistant as *T. bimaculatus* to tetraethyl pyrophosphate. It was reported by Newcomer & Dean (1949) that the Pacific mite, *Tetranychus pacificus* McG. (probably McDaniel mite, *T. mcdanieli* McG.) and the European red mite, *Paratetranychus pilosus* (C.&F.) = (*Panonychus ulmi* Koch) differed in their susceptibility to several acaricides. Garman (1950) has shown

[1] Portions of this paper represent data from a dissertation submitted by the senior author in partial fulfillment of the requirements for the Ph.D. Degree at Louisiana State University. Accepted for publication February 25, 1958.

[2] College of Agriculture, Saray Elkobba, Cairo, Egypt.

[3] Louisiana State University, Baton Rouge.

that certain colonies of greenhouse spider mites became resistant to parathion, but that resistance did not appear when several other acaricides were used.

Entomologists often utilize only the adult mite as a test animal in the evaluation of acaricides. However, Ebeling & Pence (1954) demonstrated that it is misleading to evaluate a group of acaricides on the basis of their effectiveness against a single developmental stage. They have reported that mites may vary greatly in their susceptibility to acaricides during their developmental stage. With Aramite and some of the organic phosphates they found the LD-50's for the larvae higher than for adults. In their experiments, eggs were the most resistant stage to most of the acaricides tested.

Little information is available on the relative resistance of the action of acaricides on eggs of mites, but it is often supposed that the egg is the most resistant stage. Hintz (1935) found that the stage of the European red mite most difficult to kill was the egg. He concluded that the average dosages of the organic insecticides did not prevent development of the eggs, but they often killed the larva before it could rupture the egg shell. Petty (1948) found that HETP used as a spray was not effective in killing the eggs of *T. bimaculatus* Harvey. Zimmerman & Hartzell (1947) found that the eggs of the same species were not killed in fumigation tests with HETP and TEPP in the greenhouse.

It has been demonstrated that the age of the egg may have a direct bearing on its susceptibility to most insecticides. Mukerjea (1953) determined that susceptibility varied with the age of eggs of *Tenebrio sp.*, *Diataratia oleracea*, and *Periplaneta americana*. He found that the susceptibility increased rather sharply from the 1-day egg (youngest egg) to the 2-day egg, and then gradually increased to the 5-day egg (oldest egg) which was the most susceptible.

It seems that the resistance of an arthropod egg to a toxic material may depend not only on the susceptibility of the embryonic material, but also on the ability of the toxic material to penetrate through the shell layers to the embryo. Experiments on the penetration of some types of oil through the shell layers to the embryo of several species of insect eggs have been done. In considering the ovicidal action of parathion to eggs of the peach tree borer, Smith (1955) showed that eggs treated at all stages of development continued to respire normally until the seventh day, the respiration rate declining progressively thereafter for several days beyond the normal time of hatching. Using oil saturated with a dye, Blickle (1942) found that the oil (as judged by the presence of the dye) penetrated the shell of the older eggs of tested insects more quickly than that of the younger.

Spider mites are among the more serious pests of cotton wherever it is grown. During recent years economic entomologists have been increasingly concerned with these mites, partly because many of the new insecticides are often detrimental to predaceous enemies of spider mites, or they render the plants more susceptible to the mites (Baker & Pritchard 1953).

There are at least seven species of spider mites which attack cotton in Louisiana, all of them individually capable of damaging the plant. These are: *Tetranychus telarius* (L.), *T. tumidus* Banks, *T. ludeni* Zacher, *T. cinnabarinus* (Bois.), *T. desertorum* Banks, *T. gloveri* Banks, and *T. lobosus* Boudreaux. *Tetranychus cinnabarinus*, *T. lobosus*, *T. desertorum*, and *T. tumidus* are the most widely distributed species in Louisiana. The other species seem to be of local importance only in cotton (Boudreaux 1954–55). As an example of possible severe damage to cotton, Roussel *et al.* (1951) and Roussel (1953–54) have shown that *T. tumidus* can cause serious reduction in yield (up to 77%) and reduction in length and maturity of fibers. Control of mites, therefore, may be as important to the cotton farmer as it can be to growers of other crops.

In the last few years many new acaricides have been developed. Apparently no comprehensive investigation of the relative susceptibility to these acaricides of the developmental stages of different species of mites which infest cotton has been reported in the literature. Field experience has demonstrated that some acaricides are primarily effective as ovicides, while others with practically no ovicidal effect may be highly toxic to the newly hatched young.

The objective of this study was to compare the effectiveness of four acaricides (two of which were organic phosphates), against various stages (adults, nymphs, eggs) of five species of spider mites.

MATERIALS AND METHODS.—The non-phosphate materials tested were Aramite and sulphur. The phosphate materials were Parathion and Phostex. The five species of spider mites were: *Tetranychus cinnabarinus* (Boisduval), *T. lobosus* Boudreaux, *T. tumidus* Banks, *T. gloveri* Banks, and *T. desertorum* Banks.

None of the mites tested showed any sign of being resistant. All were isolated from natural infestations. The stocks used in these experiments were reared by a method developed by Boudreaux in the entomological laboratory of the Agricultural Experiment Station at Louisiana State University (unpublished). This method permits the rapid propagation of stocks of known age.

A $\frac{3}{8}$-inch hole was bored in the center of a petri dish, and a short piece of rubber tube was fitted tightly in the hole, its upper end protruding slightly above the rim of the dish. This was placed over a plywood sheet, bored to accommodate several dishes. Below each hole in the plywood was a bottle of water. Cotton was used as the host plant, since cotton was suitable for all the species tested. When cotton seedlings were about 6 to 8 inches high, the cotyledons were nearly 2 inches across. The true leaves were not yet developed. The seedling at this stage was inserted through the rubber tube so its roots were immersed in the water below. The mites were confined to the upper surface of the cotyledon with a tanglefoot barrier. The culture was covered with a crystallizing dish 2 inches deep. The mites were easily examined without disturbance if the glass stage of binocular stereoscopic microscope was replaced with a piece of wood with a large hole cut out of the center. Such a technique allowed the microscope to rest upon a table with a cutout so that the culture dish could be placed under the microscope for examination. Large populations of mites were produced on such cultures. The cultures were maintained in the laboratory at 85 to 90° F., with fluorescent tubes furnishing about 150 foot-candles of illumination to the plants for 24 hours daily.

Pure cultures are difficult to maintain without contamination, especially in a laboratory where more than one species is cultured. The larvae apparently can be carried by air movements. It was decided, therefore, to maintain and test only one species for all the acaricides at one time. After finishing testing one species a pure culture of a different species was maintained and tested. However, cultures were examined frequently for contamination.

Method of Application of Acaricides.—The dusting chamber, gun, and air pump used in these tests were similar to those described by Waters in a publication edited by Campell & Moulton (1943), and by Ivy (1944).

Measured quantities of acaricides were calculated on a per-acre basis from the area of the dusting chamber. For instance, if dust was to be applied at the rate of 10 pounds per acre (which was used in these tests), 82 mg. of dust were required. Different concentrations of each acaricide were applied at this constant weight. A number of preliminary experiments were made to ascertain the approximate dosage range for each acaricide. The dosage mortality regression lines were based on four points. Dosages required to give mortalities of 50% were calculated from the dosage mortality relationships according to the method of Bliss (1938).

The measured quantity of each dosage was discharged from a glass gun into the top of the cylinder by means of an air pump apparatus. The dust was allowed to settle 2 minutes and the treated leaves were immediately removed. The gun was cleaned after each application by pouring clean sand into the gun, and shaking it back and forth vigorously. The sand was poured out and the process was repeated two more times.

Tests with Adults.—The petri dish method developed by Ebeling & Pence (1954) was used for all tests. Detached green cotton cotyledons were placed on wet cotton in a petri dish covered with a crystallizing dish 2 inches deep. This kept the leaves fresh for 5 days, and served also to keep all but a few of the mites confined to the leaves. It should be noted that this technique permitted fumigant action. Twenty-five adult females were placed on each treated leaf. Each experiment was replicated four times to obtain 100 mites for each dosage. Counts were made 48 hours after dusting. Mites showing very little or no movement of the legs were recorded as dead. Mites still able to crawl about, were considered alive. It should be noted here that adults made contact with the acaricides only by crawling over the residues. Untreated checks were used in each test.

Tests with Nymphs.—Ten adult females were placed on the upper surface of each leaf ringed with tanglefoot. Mites were left for 24 hours to lay an average of 60 to 100 eggs. After this period mites were removed and eggs were examined daily until all larvae had reached at least the protonymph stage. Nymphs were distinguished from larvae by the fact that the latter have three pairs of legs. Leaves were detached from the seedlings, placed on wet cotton in petri dishes, and dusted in the dusting chamber. Four applications were made of each dosage. The total number of tested nymphs for each dosage was 100 to 400. Counts were made 48 hours after treatment.

At the time the counts were made, some nymphs had already reached the adult stage, but since they had continuous contact with the residues during the nymphal stage, the experiment was considered an evaluation of the susceptibility of the nymphs. It should be noted in this test that acaricides contacted nymphs directly when applied.

Untreated controls were used for all replications.

Tests with Eggs.—For testing eggs 10 adult females were placed on the upper surface of each leaf. The mites were forced to lay eggs on the upper surface of the leaves by surrounding the leaf with tanglefoot. After 24 hours, the mites were removed from the leaves. Detached leaves were put on wet cotton in petri dishes and treated immediately. Four replications were used for each concen-

Table 1.—Summary of the susceptibilities of the various stages of the five species of mites to the acaricides tested.[a]

	Parathion		Phostex		Aramite		Sulphur	
Rank[b]	Species[c]	LD-50[d]	Species	LD-50	Species	LD-50	Species	LD-50
				Adults				
1	des.	.0064 ± .002	glov.	.094 ± .037	glov.	.130 ± .024	tum.	4.95 ± 0.590
2	glov.	.0180 ± .007	tum.	.100 ± .029	cinn.	.139 ± .004	lob.	5.00 ± 0.044
3	lob.	.022 ± .010	des.	.130 ± .020	lob.	.145 ± .020	glov.	7.40 ± 0.044
4	tum.	.0230 ± .007	cinn.	.180 ± .0004	des.	.175 ± .004	cinn.	9.2 ± 3.190
5	cinn.	.0310 ± .010	lob.	.180 ± .050	tum.	.270 ± .052	des.	10.0 ± 3.110
				Nymphs				
1	des.	.0059 ± .002	cinn.	.047 ± .012	lob.	.021 ± .002	lob.	0.68 ± .196
2	lob.	.0118 ± .002	des.	.105 ± .029	cinn.	.062 ± .017	glov.	4.50 ± 2.060
3	cinn.	.0270 ± .005	lob.	.110 ± .024	glov.	.110 ± .001	tum.	5.30 ± 1.015
4	glov.	.0320 ± .008	glov.	.200 ± .005	des.	.180 ± .0003	des.	8.40 ± 0.069
5	tum.	.0400 ± .016	tum.	.400 ± .058	tum.	.32 ± .003	cinn.	9.80 ± 2.515
				Eggs				
1	des.	.025 ± .0002	lob.	2.5 ± 0.695	lob.	2.34 ± .686	des.	48.0 ± .285
2	cinn.	.210 ± .0630	des.	2.9 ± 0.660	des.	3.00 ± .023	lob.	54%
3	lob.	.290 ± .0820	tum.	3.4 ± 1.114	cinn.	4.00 ± .832	tum.	31%
4	glov.	.350 ± .0940	cinn.	3.7 ± 0.661	glov.	4.10 ± .037	glov.	29%[e]
5	tum.	.390 ± .0770	glov.	4.2 ± 2.128	tum.	6.00 ± .020	cinn.	23%

[a] LD-50's expressed as per cent concentration of dust at 10 pounds per acre.
[b] The lower the number, the higher the susceptibility.
[c] Des. = *T. desertorum*, glov. = *T. gloveri*, lob. = *T. lobosus*, tum. = *T. tumidus*, cinn. = *T. cinnabarinus*.
[d] Differences not significant within bracketed groups. Rank within the group determined only by LD-50.
[e] LD-50 not attainable. Figures represent mortality of eggs with 100% sulphur.

tration. The number of eggs for each dose ranged from 100 to 400. Counts were made after 6 days. Partly hatched eggs, larvae, nymphs, or adults were recorded as hatched eggs, whether they were dead or alive.

Untreated controls were used for all replications. As with nymphs the acaricides contacted eggs directly when applied.

RESULTS AND DISCUSSION.—Table 1 presents the LD-50's for all acaricides tested on the developmental stages. In this table the test for significance was measured by the standard error based on the calculation of the fiducial limits from the dosage/mortality relationships according to the method of Bliss (1938). Table 2 presents a comparison of similarities and differences in the action of the acaricides on nymph and adults in such a way as to indicate possible mite relationships.

The dosage mortality curves of two species are shown in figure 1, indicating the toxicity of acaricides tested against various stages. *T. tumidus* was chosen as a representative of the most resistant species, and *T. desertorum* as a representative of the most susceptible species to the overall acaricide program.

The data of table 1 illustrate a number of phenomena: (1) Parathion was the most effective poison, Phostex and Aramite were intermediate in toxicity and about equal to each other in effectiveness, and sulphur was the least effective material; (2) the most susceptible species based on overall response of all stages seemed to be *T. desertorum*, with *T. tumidus* appearing to be the most tolerant species. *T. lobosus* approached *desertorum* in sensitivity, followed by *cinnabarinus* and *gloveri;* (3) the egg was the most resistant stage of all species with all acaricides; (4) nymph-adult comparisons indicate affinity between parathion and Phostex, placed Aramite and sulphur in classes by themselves, and grouped *tumidus* with *gloveri*, *lobosus* with *cinnabarinus* and placed *desertorum* in a group of its own. Both parathion and Phostex were more toxic to adults than to nymphs in *tumidus* and *gloveri*, but were more toxic to nymphs than to adults in the

Table 2.—Summary of relative toxicity of the compounds tested to nymphs and adults of five species of tetranychid mites.

COMPOUND	*tumidus*	*gloveri*	*desertorum*	*cinnabarinus*	*lobosus*
Parathion	A>N[a]	A>N	N>A[b]	N>A	N>A
Phostex	A>N	A>N	N>A	N>A	N>A
Aramite	A=N[c]	A=N	A=N	N>A	N>A
Sulphur	A>N	N>A	A=N	A=N	N>A

[a] This symbol indicates that adult females were more susceptible to the poison than the nymphal stages.
[b] Indicates that nymphs were more easily killed than adults.
[c] Indicates no significant difference in susceptibility between nymphs and adults.

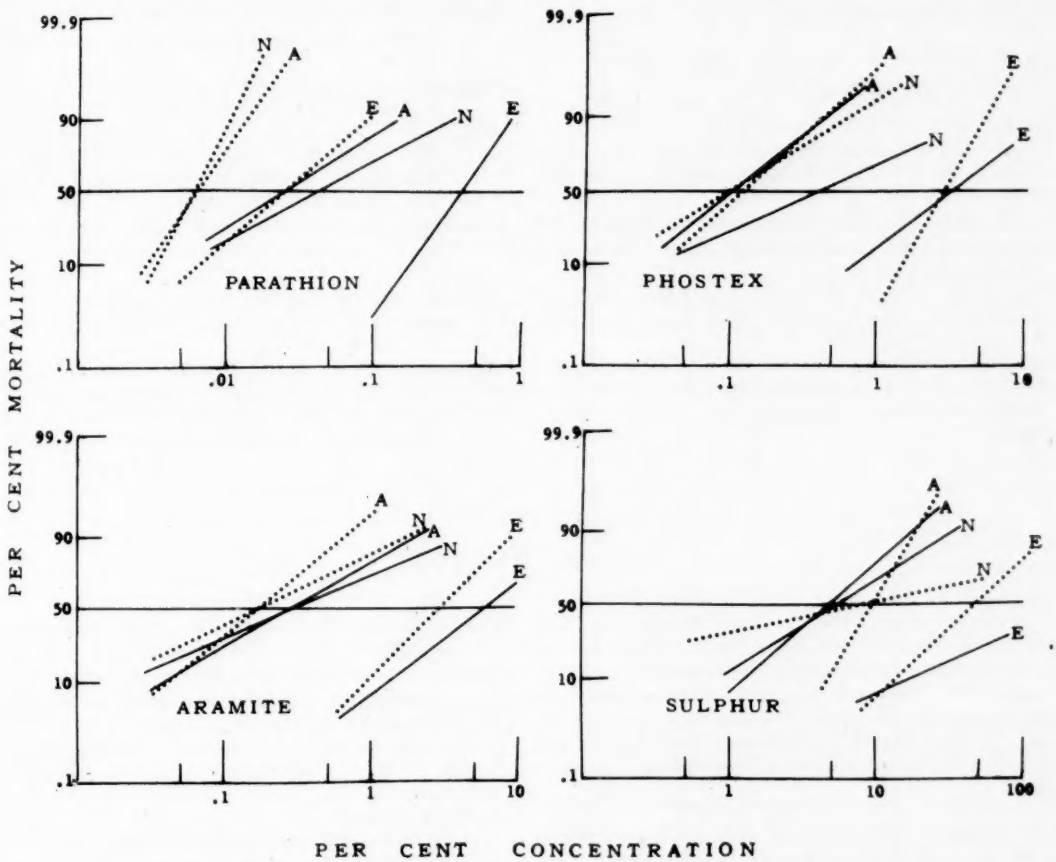


FIG. 1.—Dosage-mortality curves of *Tetranychus tumidus* and *T. desertorum*. Dotted line shows results with *T. desertorum*, solid line with *T. tumidus*. The initials represent stages of the mites as follows: A=Adults, N=Nymphs and E=eggs.

other three species. The toxicity of Aramite was about the same for adults and nymphs of *tumidus*, *gloveri*, and *desertorum*, but was greater for nymphs than for adults in *lobosus* and *cinnabarinus*. Sulphur was more toxic to nymphs than to adults in *gloveri* and *lobosus*, equal in toxicity to nymphs and adults in *desertorum* and *cinnabarinus*, and more toxic to adults than to nymphs in *tumidus*.

On a morphological basis, *tumidus* and *gloveri* are very closely related (Boudreaux 1958), *lobosus* and *cinnabarinus* are closely related (Boudreaux 1956) and *desertorum* is allied to other species of this genus (Pritchard & Baker 1953). Table 2 indicates the relative susceptibilities of nymphs to adults in such a way that these relationships become apparent. Except for responses to sulphur, *tumidus* and *gloveri* showed similar responses, and *cinnabarinus* and *lobosus* showed similar responses, but *desertorum* is not clearly similar to either group in all cases. These findings confirm the relationships of these mites as revealed by morphology.

It is apparent from these experiments that in evaluating an acaricide one must consider all stages of the life cycle. The egg stage generally was not affected by the acaricides tested when they were applied at concentrations sufficient to kill post-embryonic stages. It was also apparent that the response of one species to an acaricide cannot be depended upon to predict the response of another species.

REFERENCES CITED

Baker, E. W., and A. Earl Pritchard. 1953. A guide to the spider mites of cotton. Hilgardia 22(7): 203.

Blickle, A. L. 1942. Penetration of oils into insect eggs. (a) Influence of oil characteristics (b) Influence of age of eggs and of species. Studies of contact insecticides XVI. Tech. Bull. New Hampshire Agric. Expt. Sta. 79: 14.

Bliss, C. I. 1938. The determination of dosage-mortality curve from small numbers. Quart. Jour. Pharm. and Pharmacol. 11(2): 192.

Boudreaux, H. B. 1954–55. Summary of investigations upon cotton spider mites, Louisiana State Univ. Expt. Sta. Annual Rept. pp. 101–102.

Boudreaux, H. B. 1956. Revision of the two-spotted spider mite (Acarina, Tetranychidae) complex, *Tetranychus telarius* (Linnaeus). Ann. Ent. Soc. America 49(1): 43–48.

Boudreaux, H. B. 1958. *Tetranychus gloveri* Banks versus *Tetranychus tumidus* Banks. Ann. Ent. Soc. America 51(2): 174–7.

Campbell, F. L., and F. R. Moulton. 1943. Laboratory procedures in studies of the chemical control of insects. Amer. Assoc. Adv. Sci. Publ. 20: 115.

Ebeling, W., and Roy J. Pence. 1954. Susceptibility to acaricides of two-spotted spider mites in the egg, larval, and adult stages. Jour. Econ. Ent. 47: 879–95.

Gaines, J. C., E. E. Ivy, H. A. Dean, and A. L. Scales. 1950. Toxicity of various sulphur and phosphate compounds applied as sprays on spider mites and aphids. Jour. Econ. Ent. 45(3): 614–9.

Garman, P. 1950. Parathion resistant red spiders. Jour. Econ. Ent. 43(1): 53–56.

Hintz, H. W. 1953. Laboratory tests of acaricides on eggs of European red mite. Jour. Econ. Ent. 46: 112–5.

Ivy, E. E. 1944. Dusting device for toxicity experiments on field growth plants. Jour. Econ. Ent. 37(2): 301.

Melvin, R., and H. H. Earle, Jr. 1948. A new evaluation for acaricides using the red spider mite. Jour. Econ. Ent. 41(6): 901–3.

Mistric, W. J., Jr., and C. F. Rainwater. 1952. Comparative susceptibility of *Septanychus texazona* and *Tetranychus bimaculatus* to certain sulphur and phosphorus products. Jour. Econ. Ent. 45(1): 76–79.

Mukerjea, T. D. 1953. The relationship between the stage of development and susceptibility to DDT and pyrethrins of *Diataratia olerecea* (L.)., *Tenebrio* L. and *Periplaneta americana* (L.). Bull. Ent. Res. 44: 121–61.

Newcomer, E. J., and F. P. Dean. 1949. Comparative effectiveness of orchard acaricides. Jour. Econ. Ent. 42(5): 857–8.

Petty, B. K. 1948. Laboratory experiments with new organic insecticides. Farming in S. Africa 23: 325–32.

Roussel, J. S., J. C. Weber, L. D. Newsom and C. E. Smith. 1951. The effect of infestations by the spider mite *Septanychus tumidus* on growth and yield of cotton. Jour. Econ. Ent. 44(4): 523–7.

Roussel, J. S. 1953–54. Spider mites as pests of cotton. Louisiana State Univ. Agric. Expt. Sta. Annual Rept. p. 83.

Smith, E. H. 1955. Further studies on the ovicidal action of parathion to eggs of the peach tree borer. Jour. Econ. Ent. 48(6): 727–31.

Zimmerman, P. W., and A. Hartzell. 1947. Hexaethyl tetraphosphate and tetraethyl pyrophosphate. (I.) Their effects on plants. (II) Their toxicities to insects and mites. Contr. Boyce Thompson Inst. 15: 11–19.

BOOK REVIEW

GUIDE TO THE CHEMICALS USED IN CROP PROTECTION, third edition. By H. Martin. Dominion of Canada Department of Agriculture, Science Service. 306 pp. +9 suppl. pp. +35 pp. of index. 1957. [Looseleaf, processed.] Obtained from the Superintendent of Publications, The Queen's Printer, Ottawa, Canada. The price, payable to the Receiver General of Canada, $2.00 in Canada, United States and Mexico; $2.25 in all other countries.

The first edition of this very useful work appeared in 1951 to provide the grower, the biologist, and the chemist a guide to chemicals used for the protection of crops from insects, fungi, or weeds and an outline of their properties, characters, and analysis. The second edition was dated December, 1953, and Supplement No. 1 to this edition was issued January 1, 1955. The discovery of compounds of potential value in crop protection continues and has made necessary a complete revision of the second edition of the guide. Forty-five new compounds have been added and six compounds, now only of historical interest, have been removed. As the formulated product has now displaced, with few exceptions, the home-prepared product it has been possible to remove 18 chemicals employed to supplement the physiologically-active component in spray and dust formulations. This guide will be extremely useful to all those working with chemicals used in crop protection.

F. W. POOS

The Influence of Honey Bees on the Production of Crimson Clover Seed[1]

GEORGE H. BLAKE, JR., *API Agricultural Experiment Station, Auburn, Alabama*

ABSTRACT

Experiments were conducted in Alabama from 1951 through 1954 to determine the influence of the honey bee, *Apis mellifera* L., on the yield of seed of crimson clover, *Trifolium incarnatum* L. Colonies of bees were available in known concentrations to crimson clover fields. Observations were made during the blooming period of the clover to determine the condition of the clover plants and the number of bees visiting the clover. Cages were used to exclude the bees from small plots of blooming clover, and yields of seed from these plots were compared with those from adjacent areas to which the bees had access.

The presence of honey bees resulted in highly significant increases in seed yields of crimson clover. Under ordinary field conditions in Alabama, two colonies of bees per acre of crimson clover resulted in most efficient seed production. The average gains in total yield of seed per acre where bees were present at a rate of two colonies per acre over the yields where bees were excluded were from 281 to 509 pounds.

Where the best agronomic practices were followed, the gain in total yield of seed using three colonies of bees per acre was 1019 pounds, or a gain in yield by combine harvest of more than 800 pounds.

The number of pods formed in the clover heads was not affected by the various treatments. The percentage of the pods that contained seed was in direct proportion to seed yields. The reduction of wind or shade as caused by cages or partial cages in the clover plots had only a minor effect on yield of seed of crimson clover.

With the growth of the crimson clover (*Trifolium incarnatum* L.) seed industry and the use of crimson clover as a grazing crop in Alabama, the demand for more specific information concerning influence of the honey bee, *Apis mellifera* L., on yield of seed from this crop has increased. Numerous requests for this information prompted a quantitative study of the influence of honey bees on seed yields of crimson clover.

Alabama farmers engaged in the production of crimson clover seed as a cash crop were alarmed by the decline in seed yields from year to year. They realized that the decreasing yields might be the results of a scarcity of pollinating insects. Beekeepers were reluctant to move their bees to crimson clover fields without compensation because the blooming period of the crop is so short. The farmers were willing to pay for the service, but since very little information was available on the value of honey bees in crimson clover seed production, it was difficult to establish an equitable rental for the bees.

Literature on the value of honey bees in the production of seed of crimson clover is very scarce. Hollowell (1938) states that the florets are self-fertile, but that bees are effective in tripping the florets and transferring the pollen with a consequent increase in number of seed per head. Placing colonies of honey bees adjacent to blooming fields will effectively increase pollination of crimson clover. Beckham & Girardeau (1954) reported the results of a one-year study on pollination of crimson clover. They found that one colony of bees per acre caused the yield of seed from crimson clover to be increased by 361 pounds per acre over the yields where no bees were present. This is good general information but it indicates that more work should be done to determine the number of colonies needed for efficient seed production and the increase in yield in the presence of varying numbers of bees.

PROCEDURE.—Experiments to determine the influence of the honey bee on crimson clover seed production were initiated in 1951. Twelve cages, each 3×6 feet and 2 feet high, were constructed using one-eighth inch mesh screen wire over 2×2-inch wooden frames. The cages were used to exclude bees from small areas of clover in order that seed yields could be determined where honey bees were not a factor in pollination. Insects smaller than honey bees could pass through the screen, and the effect of exclusion of honey bees and larger pollinators could be measured.

The experiments in 1951 were conducted in three counties in Alabama. Crimson clover fields were chosen that were reported by county agricultural agents to be approximately equal in seed-yielding capacity. Colonies of bees were placed in the fields, or fields were selected that were near existing apiaries. The cages and colonies of bees were placed in the fields after grazing cattle had been removed, and at about the time the first blossoms were showing. Thus, the bees had ample time to become accustomed to the site before the pollination period began. Four cages were used at each location.

The colonies of bees used in the experiments were owned by commercial beekeepers. Each colony consisted of two to three hive bodies at the beginning of the experiments. In addition, the beekeepers agreed not to shake the experimental colonies to obtain bees for package shipment and to manage the bees so as to reduce swarming until the experiment was completed.

Bee colonies were stocked in the fields of crimson clover at rates of $\frac{2}{3}$, 2, and $5\frac{1}{3}$ colonies of bees per acre in 1951. In a 30-acre field in Lee County, 20 colonies of bees were used for pollination purposes. The colonies were placed on one side of the field on April 11 and 12. Cages were installed in the fields on April 11 and were located not more than 400 yards from the bee colonies.

In a 15-acre field in Autauga County, 30 colonies of bees (two per acre) were placed along one side of the field on April 13. The cattle were removed from the field on April 1 and the cages were installed on April 2. The cages were located from 40 to 330 feet from the bee colonies.

[1] This paper is from a thesis presented in partial fulfillment of the requirements for the Ph.D. degree in Entomology, University of Illinois. Accepted for publication March 6, 1958.

The crimson clover field in Lowndes County, also 15 acres in size, was located near existing bee colonies. One apiary site, approximately ¼ mile from the clover, contained 55 colonies, while another site, approximately ½ mile distant, contained 25 colonies. Thus, an average of 5¼ colonies of bees per acre was available for pollination of the clover.

Observations were made at intervals during the blooming period of the clover to determine differences between the clover under the cages and that outside the caged areas. Checks were also made to ensure that the bees were working in the clover fields by examining the fields for bee activity.

Yield records were taken from May 19 to May 23, 1951. The areas under the cages (18 square feet) were harvested to determine seed yields in the absence of honey bees, while areas of the same size near the cages were harvested to determine the seed yields where bees were present. In order to keep shattering of ripened heads to a minimum, the plots were harvested by hand and all of the foliage was collected. The stems were clipped just above ground level with handshears and the plants were placed in cloth bags. The seed was separated from the foliage by a miniature threshing machine and cleaned with a Clipper cleaner. Yields, in grams of clean seed per plot, were determined and were later converted to pounds of seed per acre.

In 1952, the experiments were basically the same as those in 1951. However, certain observations during the previous season led to additional investigations. The experiments were conducted at four locations in three counties. In three of the experiments, treatments were the same as in 1951.

Two fields in Lowndes County were selected for use in 1952. In one of the fields, two colonies of bees per acre were available for pollination and in the other approximately 10 colonies per acre were available. The latter figure is an approximation, since 66 colonies plus 2700 nuclei were available. Ten nuclei are about equal to one colony.

A third experiment was conducted on the Station's Agricultural Engineering Unit at Auburn. In this experiment, the best known agronomic practices were used to obtain a maximum seed yield. The clover was irrigated as needed and was not grazed. Three colonies of bees per acre were placed in the field. The total seed yield as determined when the clover was harvested by hand was compared with the yield obtained when the clover was harvested by commercial combining.

A fourth experiment was begun in Montgomery County to determine the effects of shading and the reduction of wind in the caged areas. Four cages were used to enclose the clover and exclude the honey bees (tops and sides, bees excluded); four cages were used without tops to reduce the wind (sides only, bees not excluded); and four cages were used with the sides extended horizontally to shade the clover (tops only, bees not excluded). Colonies of honey bees at a rate of 1½ colonies per acre were centrally located in the field.

In 1953 and 1954, all of the cages were used in one crimson clover field. To the four treatments used in Montgomery County in 1952, another was added in which areas of clover were enclosed but bees were able to enter the caged area (caged, bees not excluded). In order to permit entry to the caged area, each was attached to a hive and a hole was cut in the screen adjoining an opening in the hive. The opening in the hive was provided by moving the supers above the brood chamber (first hive body) backward about 1½ inches. This left a similar opening in front of the hive and to prevent water from entering the brood chamber a small board was secured in place over the opening. The regular entrance to the hive was left open enabling the bees to work either the clover in the cages or that outside. Once in the caged area, however, the bees had to return to the hive to reach the outside clover.

One and one-half colonies of bees per acre of crimson clover were provided in 1953 and 1954. Each treatment was replicated six times in 1953 and five times in 1954.

One bee count on April 17 and two counts on April 23, 1953 were made to determine the number of bees visiting the clover in the different areas. Counts were made simultaneously for all treatments in each replicate to eliminate variation due to time of day.

Yield records were obtained in 1953 as described previously, but in 1954 the seed was stripped from the plants by hand. In 1953 the yield obtained by combine as well as the total yield was determined. Just prior to harvest in 1953 and 1954, 10 clover heads were collected from each plot in the experiment. The number of seed pods formed on the heads, the number of these pods containing seed, and the percentage of the pods containing seed were determined.

The effect of distance was not determined in this study, since the fields were relatively small and, in most instances, the colonies were placed along the edges of the fields or in the fields in positions to require a minimum flight. In all of the experiments, fields were selected that were no closer than 1 mile to other crimson clover fields, and since the main bloom of white clover, vetches, and other competitors for pollinators are generally just beginning to bloom when the crimson clover bloom is at its height, the bees had to work the test field or fly a considerably greater distance to obtain nectar.

RESULTS AND DISCUSSION.—In all of the 1951 experiments, pronounced differences were observed in early May between the clover in the caged areas and that outside the cages. The heads of clover under the cages were still in full bloom, while the heads of clover outside the cages had either stopped blooming or were blooming at the tips only. The latter heads had already set a crop of seed. The foliage of the caged plants was also greener than that of clover outside the cages. These differences were observed in all subsequent experiments.

The average yields of seed in the three experiments in 1951 are presented in table 1. When the average gains in seed yields are studied in relation to the number of colonies of bees per acre, a positive correlation is evident. The relationship is shown in figure 1.

Further evidence of the relation between seed production and the number of colonies of bees available per acre was obtained in 1952 and is presented in tables 2 and 3. The data in table 1 and in columns 1 and 3 of table 2 show that, under ordinary field conditions, two colonies of bees per acre of crimson clover result in most efficient seed production. The data in the center column of table

Table 1.—Yields of crimson clover seed in the absence or presence of honey bees in varying numbers of colonies per acre. 1951.

Treatment	Average Yield[a] at Indicated Number of Colonies of Bees per Acre		
	⅔	2	5⅓
Bees excluded	156.5	92.1	40.3
Bees present	540.6	600.7	575.2
Average gain (lbs.)	384.1	508.6	534.9
L.S.D. at 1% level	230.5	338.7	431.5

[a] Mean of four replications.

2 reveal that, where the potential yield of seed is raised by following the best agronomic practices, a greater number of bees per acre causes a correspondingly greater increase in seed production. It is interesting to note that in this experiment, where three colonies of bees per acre were present, the average yield of honey gathered from the clover over a 2-week period was slightly greater than 100 pounds per colony. Normally, crimson clover will yield nectar for more than 3 weeks. One colony of well-managed bees will gather from 76 to 106 pounds of honey during this period (Girardeau 1954). In 1952 adverse weather conditions for the first week limited bee activity to 2 weeks, yet the bees gathered a normal amount of honey.

The data presented in table 3 help explain the effects of shading and the reduction of wind in the caged areas.

The presence of bees, regardless of the treatment in other respects, resulted in highly significant increases in yields when compared with yields where bees were excluded. There were also significant differences among the

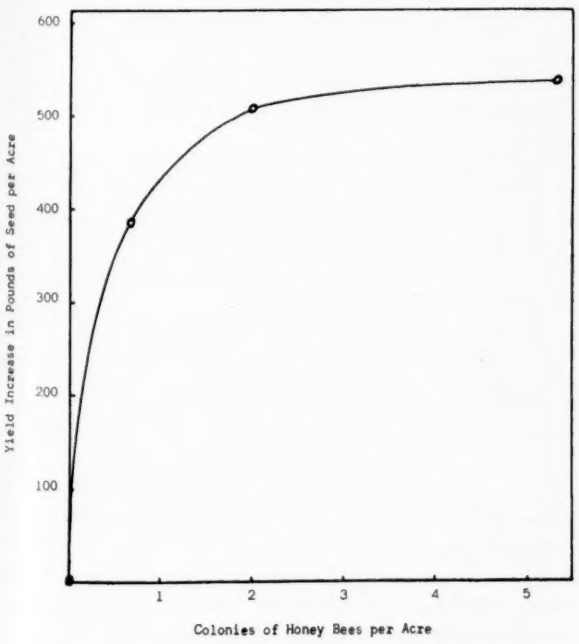


FIG. 1.—Relationship of increase in seed yield of crimson clover to availability of honey bee colonies.

Table 2.—Yields of crimson clover seed in the absence or presence of honey bees in varying numbers of colonies per acre, 1952.

Treatment	Average Yield[a] at Indicated Number of Colonies of Bees per Acre		
	2	3[b]	10
Bees excluded	19.4	176.6	49.0
Bees present	300.2	1,195.8[c]	308.5
Average gain (lbs.)	280.8	1,019.2	295.5
L.S.D. at 5% level	209.0	—	146.0
at 1% level	—	283.5	—

[a] Mean of four replications.
[b] Best known agronomic practices were followed.
[c] Average yield combined was greater than 800 pounds per acre.

Table 3.—Seed yields of crimson clover in the absence of honey bees and when bees were available at 1.5 colonies per acre under certain conditions, 1952.

Treatment	Average Yield[a] in Pounds per Acre
Bees excluded; wind reduced; shaded	54.3
Bees not excluded, wind reduced	409.5
Bees not excluded, shaded	546.0
Bees not excluded, open	938.7
L.S.D. at 5% level	272.0
at 1% level	189.2

[a] Mean of four replications.

treatments in which bees had access to the clover. There was no significant difference in the effects of wind reduction or shade. The treatments with partial cages caused reductions in yields that were highly significant when compared with yields where the clover was not caged.

In 1953, information was obtained on the number of bees visiting clover subjected to the various treatments. The results of the bee counts reported as averages of the six replications are presented in table 4. An occasional bumblebee was observed in the experimental fields, but populations of bumblebees and other bees were so small that their effect on pollination was deemed insignificant.

Table 4.—Number of bees visiting 1 square yard of crimson clover over a period of 1 hour when bees were available at a rate of 1.5 colonies per acre, 1953.

Conditions with Bees not Excluded	Average Number of Bees at Designated Time[a]			Overall Average (bees/ sq. yd.)
	April 17 (noon)	April 23 (A.M.)	April 23 (P.M.)	
Wind reduced; shaded	32	44	43	39.7
Wind reduced	6	19	18	14.3
Shaded	19	40	26	28.3
Open	41	32	37	36.7
L.S.D. at 5% level				29.1
at 1% level				38.6

[a] Average of six replications.

Table 5.—Number of seed pods per head of crimson clover and per cent of pods containing seed in the absence or presence of honey bees under certain conditions, 1953[a] and 1954.[b]

CONDITIONS	AVERAGE NUMBER OF SEED PODS/HEAD	PER CENT OF PODS CONTAINING SEED	ANGLES	POUNDS OF SEED PER ACRE
	1953			
Bees excluded, shaded and wind reduced	87.6	11.2	19.31	140.7
Bees not excluded, shaded and wind reduced	92.7	74.0	59.92	425.4
Bees not excluded, wind reduced	91.7	77.7	61.87	627.6
Bees not excluded, shaded	101.6	76.7	61.13	722.9
Bees not excluded, open	97.0	83.3	66.04	830.2[c]
L.S.D. at 5% level	n.s.	—	5.38	183.4
at 1% level	n.s.	—	7.35	250.7
	1954			
Bees excluded, shaded and wind reduced	84.7	18.9	25.78	46.6
Bees not excluded, shaded and wind reduced	73.1	58.9	50.12	165.2
Bees not excluded, wind reduced	69.4	53.3	46.88	92.5
Bees not excluded, shaded	66.2	53.5	47.02	100.7
Bees not excluded, open	60.0	68.1	55.61	91.0
L.S.D. at 5% level	n.s.	—	13.05	66.32
at 1% level	n.s.	—	19.29	—

[a] Average of six replications.
[b] Average of four replications, except seed yields were based on five replications.
[c] Combined seed yielded 385 pounds per acre.

When the population counts were made, no bees other than honey bees were observed in the field.

Observations in 1953 revealed that, in the treatment where the clover was caged with bees having access to the caged clover, the bees would enter the caged areas and work the clover blossoms in a normal manner when the hive opening into the cage was facing north. When the hives were reversed, many bees, upon entering the cage, became confused when they confronted the screen and were killed. In 1954, therefore, all hive openings into the cages of bee colonies used for this treatment were placed towards the north.

There were no significant differences among the treatments exposed to bees although the number visiting the areas in which wind was reduced was considerably lower than in the other plots where bees were available.

The results of 1953 and 1954 studies made to determine if the differences in seed yields of crimson clover from the various treated plots were due to number or weight of seed formed are given in table 5. There were no differences in the number of pods formed in the clover heads, but the percentage of the pods containing seed varied to a highly significant degree. In all of the treatments where bees were available, a much greater percentage of pods contained seed than where bees were excluded. In 1953 there were also differences in the percentages of pods containing seed among the treatments to which bees had access.

The total yields for the treatments in 1953 and 1954 are given in table 5. The average yield obtained by commercial combine is also presented for 1953. In 1954, only the plot areas were harvested.

In 1953, as in 1952, the presence of honey bees, regardless of treatment in other respects, resulted in highly significant increases in seed yields of crimson clover, when compared with yields where bees were excluded. The 1954 season was extremely dry; as a result, the yields of seed were low. In that year, only in one treatment where bees had access to clover (bees not excluded, shaded and wind reduced) were the yield increases significantly greater than those where bees were excluded. The other treatments where bees were available, however, approached significance. Although the differences in yield among treatments to which bees had access were not significant, the caged areas with bees present produced more seed than the other areas to which bees had access. The relatively higher yield in the areas caged with bees present in 1954, can probably be attributed to the presence of more moisture in caged areas, since moisture was a limiting factor in seed production.

A comparison of the two caged treatments in 1953 shows that the presence of bees resulted in an average increase in seed yield of 285 pounds per acre. This increase was the actual benefit derived from the bees, since all variables, except presence or absence of honey bees, should have been the same, There was, however, no way of knowing what the benefit of the bees would have been, had the clover not been caged.

Differences in maturity of clover in the areas from which bees were excluded and the areas where bees were available, irrespective of treatments, were evident in all of the experiments. Most pronounced differences were observed between areas caged with no bees present and uncaged clover.

Clover in plots that were shaded or plots in which wind was reduced was slightly later in maturity than the uncaged clover, as evidenced by differences in color of the foliage and in the development of the heads. Observations in 1952 and the counts of bees in 1953 revealed that fewer bees visited the areas that were shaded and the areas in which wind was reduced. Wind reduction was not a factor in the visitation by bees, however. As they drifted from clover blossom to clover blossom, the bees would be confronted by the sides of the cage and would fly up, and in most instances, over the enclosed area. Other bees, however, would fly directly to the enclosure and visit the blossoms in a normal manner. The bees

would readily enter the shaded areas, but generally the visits were shorter than normal.

In Montgomery County in 1952, the data on seed production seemed to indicate that wind reduction or shade, as such, had a considerable influence on the seed yields. Actually, only a small amount of the reduction in yield is caused directly by these two factors. Their greatest effect is on the bee population, as is indicated in the preceding paragraph, since fewer bees or shorter bee visits in the areas caused the reductions in yield.

REFERENCES CITED

Beckham, C. M., and J. H. Girardeau, Jr. 1954. A progress report on a study of honey bees as pollinators of crimson clover. Georgia Agric. Expt. Sta. Mimeo. Ser. **70.** 8 pp.

Girardeau, J. H., Jr. 1954. Reseeding crimson clover as a major honey plant in South Georgia. Georgia Agric. Expt. Sta. Mimeo. Ser. (N.S.) **1.** 7 pp.

Hollowell, E. A. 1938. Crimson clover. U. S. Dept. Agric. Leaf. 160. 8 pp.

Walnut Aphid Investigations in Northern California[1]

A. E. MICHELBACHER and STEPHEN HITCHCOCK, *University of California, Berkeley*

ABSTRACT

The walnut aphid (*Chromaphis juglandicola* (Kltb.)) is most destructive to walnut from the time the leaf buds begin to unfold until the leaves are fully expanded. Severe infestations during this period greatly stunt the leaves which is reflected in a reduced size of the developing nuts.

Schradan is the most effective insecticide that has been tested against the walnut aphid. Except in a region where the aphid has shown resistance to certain phosphate insecticides, 0.5 pound of schradan per acre has effectively controlled the pest for an entire year. Better control has been obtained with dosages of 0.75 and 1.0 pound. For satisfactory results treatments must be made before the foliage hardens. Other factors that influence the effectiveness of treatment are vigor, and thoroughness of application.

Although schradan exerts a suppressing action on other sucking pests of walnut, there are some cases, such as the calico scale (*Lecanium cerasorum* (Ckll.)) and the European red mite (*Panonychus ulmi* (Koch)), where it tends to induce an increase in the population.

The vigor of the trees and the exceptional fine quality of foliage on trees treated with schradan may in part result from a stimulus beyond that obtained through the control of the walnut aphid and other pests.

Because it is ever present the walnut aphid, *Chromaphis juglandicola* (Kltb.) commands continual attention. The status of the pest through 1954 has been reported upon by Michelbacher (1955). Since that time much additional information concerning the insect has been obtained.

A more exact evaluation of the aphid's economic position has been made possible through the remarkable suppressing action of schradan (octamethylpyrophosphoramide) upon the pest. The early results obtained with this systemic insecticide have been reported upon by Michelbacher & Oatman (1955). It was shown that a single treatment applied in early May resulted in nearly perfect control of the walnut aphid for a period of 1 year. Since then, further extensive investigations with the insecticide have been conducted and some plots have received yearly treatments over a 4-year period. The experimental studies were directed towards determining the most effective time to apply the insecticide and the lowest dosage that would result in effective control. Further, observations were made on the action schradan exerted on other pests of walnut. The investigations were conducted in several localities in northern California, which included orchards at Linden, Modesto, Walnut Creek and San Jose.

Populations of the walnut aphid were determined by counting all individuals on the next to the terminal leaflet. Treatments were usually replicated twice, and most plots consisted of 80 trees, or approximately 4.5 acres. All applications were made with air carrier sprayers in 100 to 200 gallons of water per acre. Unless otherwise noted, the investigations were conducted on the Payne walnut, a variety that comes into leaf early in the season.

1955 INVESTIGATIONS.—An unusually heavy infestation of the walnuut aphid was encountered in early season throughout most of the walnut orchards in the San Joaquin and Sacramento Valleys. The leaves on the early varieties were seriously attacked as soon as the buds began to unfold in the spring. The severe infestation resulted in a stunting of the developing leaves. By May 1, the average number of aphids in some orchards exceeded 50 per leaflet. The beneficial action of year-long protection afforded by schradan was well illustrated in the experimental orchard at Linden. In table 1 are given the aphid treatments and the aphid population prior to treatment, and the population encountered on the last survey made on October 28. It should be noted that the plots that received schradan treatments in 1954 were nearly free of aphids although a year had elapsed since the sprays were applied. The foliage on these trees was large and in beautiful condition when compared with plots where aphid treatments had not yet been applied. In some of the latter the leaves were hardly half as large and appeared sickly when compared with those from plots that had received schradan in 1954. Further, the foliage was dripping with honeydew. A better demonstration of destructiveness inflicted by the aphid in early season could not have been found.

[1] Accepted for publication March 10, 1958.

Table 1.—Walnut aphid populations in 1955 prior to treatment, and in late season at Linden, California, showing the outstanding action of schradan against the pest.

Aphicide and Pounds per Acre		Average Number of Aphids per Leaflet on: May 4	Oct. 28
Demeton (actual)[a]			
0.25		43.0	5.1
0.50		40.0	4.3
0.75		30.4	5.8
Schradan (actual)[a]			
0.55		31.5	0.3
0.78		35.2	0.0
1.17		0.6[b]	0.0
1.56		0.3[b]	0.0
Parathion 25%, w.p.	1.0[c]	52.0	6.5
Malathion 25%, w.p.	3.0[c]	65.9	7.0
Nicotine 25% dry concentrate	5.0[c]	45.7	9.0
BHC 12% gamma isomer[d]	3.75	43.9	3.1

[a] Applied May 12, 1955.
[b] Schradan applied in 1954.
[c] Two applications May 14 and July 1.
[d] Applied May 14, followed by 0.25-pound dosage of demeton on July 1.

In the 1955 investigations an attempt was made to determine the lowest dosage at which schradan would give satisfactory control. At Linden it was applied at rates that ranged from approximately 0.5 to 1.5 pounds per acre. The information in table 1 shows that good results were obtained at all dosages, although at the lowest level there were more aphids present in late season than where the larger amounts were applied. Similar results were obtained in the experimental orchards at Modesto and Walnut Creek. Although excellent, the suppressing action of schradan in the experimental orchards at San Jose did not quite equal the performance obtained in the other localities. The San Jose area is a region where the walnut aphid was found to be resistant to parathion (Michelbacher *et al.*, 1954). This fact may account for the type of response to schradan encountered in the latter area.

Investigations have shown that in the early season the walnut aphid is not as destructive to the late varieties as it is to the early ones. This is due to the fact that the foliage usually is nearly fully expanded before a destructive aphid population has had an opportunity to develop. In order to determine the effect of the aphid upon a late variety a block of Franquettes was sprayed with 0.8 pound of schradan per acre on May 16 when the nuts were tiny and in the feather stage. In this block not a single aphid was found in 10 surveys conducted from May 24 through September 21. The aphid population in surrounding blocks reached a point where the trees dripped with honeydew, and the foliage and the developing nuts became blackened due to the growth of sooty mold fungus. At harvest the plot sprayed with schradan was green and held its leaves while the surrounding trees were nearly defoliated. Despite the marked contrast, the yield and quality of nuts from the schradan-sprayed trees, although better, was not greatly superior to that of adjacent blocks. One of the greatest benefits derived from the treatment was that the trees held their leaves which greatly facilitated harvest operations. This same block of treees was similarly treated with schradan in 1956 and 1957. The results were similar to those obtained in 1955. There was some increase in yield and quality of nuts, and the fact that the trees held their leaves again simplified harvest operations. It is also interesting to note that the treated block of trees remained practically free of aphids for the entire 3-year period.

1956 Investigations.—Studies with schradan were largely directed towards obtaining further information on the lower effective dosage levels. In addition some experiments on timing of applications were conducted, and the possible effect of schradan on other organisms was observed. The early season aphid population in the central valley, as in 1955, was very large. On early varieties serious damage occurred where control measures were neglected or were inadequate. In the experimental orchard at Linden schradan was applied at rates that ranged from approximately 0.4 to 1.5 pounds per acre. The earliest treatments were applied on April 17 and the remainder of the treatments at the end of April and the first of May. At the time the latter treatments were applied the leaves were completely unfolded and the average cross-sectional diameter of the developing nuts ranged from $\frac{3}{8}$ to $\frac{1}{2}$ inch. The treatments used and the degree of control obtained are shown in table 2. The outstanding effectiveness of schradan is apparent when the results are compared with those obtained with other aphicides. The carry-over influence of schradan is clearly shown where it had been applied the previous year. It should also be noted that some relation existed between the dosage applied and the degree of control obtained. In general the protection secured increased with dosage. The April 17 applications were made to plots that received schradan treatments for the first time. No detectable damage from the aphid in these plots was apparent even though the aphid population averaged 20 per leaflet at time of treatment.

The pre-treatment early season trend of the aphid population in the plots receiving demeton paralleled that obtained with schradan. The explanation for this is that a serious soft scale population had developed in these plots and they were treated on February 16 with 25% parathion wettable powder, at the rate of 4 to 5 pounds per acre, to control the scale. The treatment was also highly effective in destroying the overwintering eggs of the walnut aphid.

Results similar to the above were obtained in the experimental orchards at Modesto, Walnut Creek and San Jose. However, in the latter area the results with schradan did not quite equal those obtained in the other areas.

The results obtained with schradan in the experimental orchard at Modesto are shown in table 3. The 0.43-pound treatment gave better control than the 0.6-pound dosage. The probable reason for this is that the latter plot had received demeton treatments during previous years and was heavily infested with soft scales. The scale population seriously interfered with the vigor of the trees, and they were in poor condition when compared with the plots that had received schradan in the prior year. The reduction in vigor apparently adversely influenced the absorption of schradan. Numerous other observations were made that strongly indicated that vigor and age of foliage

Table 2.—Relative effectiveness of several aphicides in controlling the walnut aphid at Linden, California, 1956.

APHICIDE, POUNDS PER ACRE, AND DATES OF APPLICATIONS	AVERAGE NUMBER OF APHIDS PER LEAFLET ON: Apr. 5	Apr. 17	Apr. 24	Apr. 30	May 8	June 14	June 20	July 3	Aug. 3	Sept. 5	Sept. 14	Oct. 5
Demeton 0.25, May 2 (actual)	0	0	3.6	—	0	0	—	0.1	3.0	16.7	14.9	2.5
0.25, May 2 and Sept. 5	0	0	6.8	14.1	0	0	—	0	0.8	28.4	0.3	4.9
0.50, May 2	0.1	0.1	1.6	—	0	0	—	0	0.6	22.3	11.0	4.3
Schradan 0.40, Apr. 17 (actual)	5.4	20.5	0	—	0	0	—	0	0.1	0.5	2.1	1.1
1.00, Apr. 17	6.7	20.5	0	—	0	0	—	0	0	0.2	0	0
0.55, Apr. 30[a]	0.6	1.3	10.7	16.0	0	0	—	0	0	1.0	3.6	1.6
0.78, Apr. 30[a]	0.3	0.4	7.2	9.7	0.1	0	—	0	0	0.4	0	0.5
0.80, May 1[a]	0	0	1.8	—	0	0.1	—	0	0	0	0	0
1.17, May 1[a]	0	0.1	0.7	1.6	0	0	—	0	0	0	0	0
1.56, May 1[a]	0	0.1	1.2	3.8	0	0	—	0	0	0	0	0
Parathion 25%, W.P. 1.0 Apr. 19 and June 20	11.0	41.5	0.1	0.1	0	2.1	13.3	0	0.1	29.3	25.2	1.8
Malathion 25%, W.P. 3.0 Apr. 19 and June 20	13.0	52.7	0	0.3	0.1	7.4	28.8	0	0	19.6	50.8	1.9
Nicotine 25%, W.P. 5 Apr. 24 and June 20	—	39.7	87.8	0.2	0	6.1	15.4	0	0.4	21.4	24.5	2.6
BHC, 12% gamma isomer 3.75 Apr. 19 and Demeton 0.25 June 20	4.8	27.6	0	0.5	0.1	16.5	31.1	0	0	1.0	7.0	0.5

[a] Schradan applied to these plots in previous years.

influence the effectiveness of schradan. Poor control resulted where growers applied treatments to trees after the foliage was mature. Thomas (1956) reported that appreciable amounts of scharadan were still present on the outer surface of apple leaves 39 and even after 60 days when applications were made at the beginning of September.

It is possible that much of the chemical is washed from the foliage when rains closely follow treatments applied to foliage after it has hardened. Such action may have accounted for the poor control experienced by a grower where a heavy rain occurred the same day he applied schradan to trees with maturing foliage. Rain within a day after treatment has not apparently interfered with the effectiveness of experimental treatments where the application has been made to young foliage.

That vigor is important is supported by the investigations of Fjelddalen (1955) who reported that schradan was most effective when applied to young plants which are growing vigorously. Others who have stated or indicated that schradan is more readily absorbed by young foliage include Ripper *et al.* (1950, 1951), Jefferson (1951) and Zeid & Cutkomp (1951).

Table 3.—Control of walnut aphid obtained with various dosages of schradan applied April 25, 1956 at Modesto, Calif.

POUNDS PER ACRE	AVERAGE NUMBER OF APHIDS PER LEAFLET ON: May 14	June 11	July 20	July 31	Aug. 13	Aug. 30	Sept. 29	Oct. 20
0.43[a]	0	0.1	0	0	0	0	0.5	0.1
0.6[b]	0	0.2	0.1	0.1	0.1	1.5	1.9	1.3
1.0[a]	0	0	0	0	0	0	0	0
2.1[a]	0	0	0	0	0	0	0	0

[a] Treated with schradan previous year.
[b] Plot heavily infested with soft scales.

1957 INVESTIGATIONS.—The principal studies with schradan were directed towards determining the earliest period that it could be effectively applied to walnuts to control the walnut aphid. At Linden the dosage used ranged from 0.5 to 1 pound per acre and applications were made over a period that ranged from April 3 through April 30. On April 3, the most advanced walnuts had just reached the feather stage, and the leaves were extremely succulent but very small. By April 30 the average cross-sectional diameter of the walnuts was $\frac{3}{8}$ to $\frac{1}{2}$ inch. The leaves were fully expanded, but still in a succulent condition.

The results along with those obtained with other aphicides are given in table 4. Regardless of the time they were applied, the schradan treatments were much more effective than any of the other aphicides. There is some evidence that the control obtained with the earliest applications was not quite as impressive as that resulting from the later treatments. While the suppressive action obtained with the 0.5 pound treatment was good, the trees that received the 0.75 pound and 1 pound applications looked the best. The favorable comparison in early season of the BHC-demeton treatment with the schradan treatments is due to the fact that the overwintering aphid eggs were killed when the former plots were treated in November with parathion to control soft scales.

Of the new materials tested against the aphid, Trithion, ethion, and Guthion showed a great deal of promise. This is particularly true if they are compared with the results obtained with parathion and malathion.

At Walnut Creek excellent control of the walnut aphid was obtained with schradan at a rate as low as 0.4 pound per acre. At San Jose a single application of schradan was applied at 0.5 pound per acre. At the time of applica-

Table 4.—Relative effectiveness of several aphicides against the walnut aphid at Linden, California, 1957.

Aphicide, Pounds per Acre, Dates of Applications	Average Number of Aphids per Leaflet on: Apr. 2	Apr. 16	Apr. 23	May 7	May 24	June 12	June 20	July 23	Aug. 26	Sept. 9	Sept. 17	Oct. 9
Demeton 0.25, Apr. 29 and June 26[a] (actual)	2.0	15.2	20.0	0	0.1	0	0.1[b]	0	0	2.1	6.4	36.5
0.50, Apr. 29[c]	1.3	15.7	20.1	0	0	0	0[b]	0	0.9	17.7	20.0	21.0
Schradan[d] 0.50, Apr. 3[e] (actual)	0.6	1.3	0.6	0	0	2.0	0.5[b]	0	0	1.0	1.7	16.6
0.50, Apr. 9[e]	1.0	0.6	0.1	0	0	0	0[b]	0	0	0.1	0.2	7.0
0.50, Apr. 16[e]	0	0.8	0.1	0	0	0	0[b]	0	0	0.2	0.8	6.5
0.50, Apr. 23[e]	0	0.2	1.6	0	0	0.1	0[b]	0	0	0.2	0.4	0.8
0.50, Apr. 29[c]	0.4	3.5	8.4	0	0.1	0	0.3[b]	0	0	1.2	1.0	11.4
0.75, Apr. 23[e]	0	0.1	1.2	—	0	0	0[b]	0	0	0.0	0.1	0.2
0.75, Apr. 30[e]	0.1	1.4	5.0	0	0	0	0[b]	0	0	0.1	0.1	1.1
1.00, Apr. 3[e]	0	0	0	0	0	0.2	0.1[b]	0	0	1.1	0.3	1.3
1.00, Apr. 9[e]	0.2	0.1	0.1	0	0	0	0[b]	0	0	0.1	0	0.9
1.00, Apr. 16[e]	0.2	0.7	0	0	0.1	0	0[b]	0	0	0	0	0.4
1.00, Apr. 30[e]	0	0.1	0.5	0	0	0	0	0	0	0	0.2	0.6
Parathion, 25% W.P. 1; Apr. 26 and June 26[b]	1.8	8.1	15.8	0	0	4.8	10.4	0	0.4	10.4	25.0	38.6
Malathion, 25% W.P. 1; Apr. 26 and June 26[b]	2.4	13.0	18.4	0	0	1.9	2.7	—	0.1	5.0	19.5	31.3
BHC, 12% g. i. 3.75, May 1 and Demeton 0.25, June 26[b/f]	0.1	1.2	1.8	0	0	0.5	0.6	0	0	1.1	4.4	32.2
Guthion, 15% W.P. 6.64; Apr. 23 and 5.6 June 21[b]	1.9	10.5	13.4	0	0	7.6	16.7	0	0	1.1	3.1	26.1
Ethion, 25% W.P. 10, Apr. 26 and 15, June 21[c]	1.8	7.2	7.8	0	0	0	0.2	0	0	0.4	4.1	28.5
Thiodan, 25% W.P. 10, Apr. 26 and 15, June 21[c]	2.1	13.4	26.4	0	0	0.5	0.2	0	0.1	1.1	2.3	26.3
Trithion, 4 pounds per gallon, 1 qt. Apr. 16 and June 26[c]	—	15.0	0	0	0	0.7	0.3	0	0	0.7	3.7	31.2

[a] Early treatment applied in 200 gallons of water per acre, 2nd in 100 gallons of water.
[b] Plots treated with 2 pounds 25% parathion W.P. on June 26, 1957 to control soft scales.
[c] Applied in 200 gallons of water.
[d] All plots treated with schradan in previous year.
[e] Applied in 100 gallons of water.
[f] Treated with parathion and oil November 23, 1956.

tion (April 11) tree growth was vigorous although the leaves were still very small. The control obtained was poor when compared with that secured in other areas. On July 9 the average aphid population per leaflet had reached 4.1 and the following day the plot was retreated with a different aphicide. The results obtained with schradan at San Jose in 1957 and the preceding 2 years are given in table 5. Except where large dosages of schradan were used the results do not compare favorably with those obtained in other areas. Although age of foliage, vigor of growth and type of equipment used may have exerted some influence, it is believed that the relatively poorer results obtained at San Jose were associated with the fact that the walnut aphid in that region is known to be somewhat resistant to certain phosphate insecticides.

FACTORS INFLUENCING EFFECTIVENESS OF SCHRADAN.—Extensive investigations with schradan since 1953 have shown that its effectiveness against the walnut aphid is dependent upon a number of factors. Important among these are age of foliage, vigor of growth, and thoroughness of coverage. The experimental information clearly indicates that satisfactory results depend upon applications being made before the foliage hardens. Good control was obtained from the time the leaf buds started rapid growth until the leaves were fully expanded. There is certainly more danger of applying schradan too late than too early. Except in the region where the walnut aphid has demonstrated resistance to some phosphate insecticides, satisfactory control has been obtained with dosages of schradan as low as 0.5 pound per acre. Thoroughness of application is extremely important. On some occasions the equipment used failed to apply the spray thoroughly to the top of large trees. In such cases the upper reaches of the trees became heavily infested, and dripped with honeydew. In contrast, the lower portions remained free of the pest for the entire season.

INFLUENCE ON OTHER PESTS.—As used against the walnut aphid schradan appeared to have exerted a suppressing action on the frosted scale (*Lecanium pruinosum* Coquillett)-European fruit lecanium (*L. corni* Bouché) complex, but not on the calico scale, *L. cerasorum* Ckll. With the latter schradan actually appeared to stimulate

Table 5.—Control of the walnut aphid with applications of schradan at San Jose, Calif., during 1955–57, inclusive.

	Pounds of Active Ingredient per Acre	Date Applied	Average Number of Aphids per Leaflet									
			May		June		July		Aug.		Sept.	
			1–15	16–31	1–15	16–30	1–15	16–31	1–15	16–31	1–15	16–30
						1955						
Orchard A	1.10	May 18	—	—	0	—	0.7	2.1	—	2.2	0.7	0
	1.46	May 18	—	—	0	—	1.2	0.1	—	0	0	0
Orchard B	1.15	May 26	—	—	0	—	0	0	—	0	0	0
	2.77	May 26	—	—	0	—	0	0	—	0	0	0
						1956						
Orchard A	0.87	May 15	4.2[a]	0.1	0	—	0	0.2	0.4	0.9	1.2	1.5
	1.12	May 18	0.6	0	0.1	—	0	0	0	0	0.1	0.2
Orchard B	1.08	May 22	—	9.1	0	—	0.1	0.1	0.1	0	0.1	0.2
						1957						
Orchard B	0.50	Apr. 11	0.3	1.7	0.1	1.04	4.1	—	—	—	—	—

[a] Pretreatment count.

an increase in the population. The degree of the suppressive action on the frosted scale-European fruit lecanium complex probably increased with dosage. However, no beneficial results could be detected in cases where the orchards or plots were heavily infested at the time the treatment was applied.

Little is known about the action of schradan against armored scales. The Putnam scale, *Diaspidiotus ancylus* (Putnam) increased to a somewhat destructive level in plots at Linden which had received three yearly treatments of schradan at 1 pound or more per acre.

Good suppression of the Pacific spider mite, *Tetranychus pacificus* McG. and the two-spotted spider mite, *T. telarius* (L.) was obtained with all dosages of schradan. This was not the case with the European red mite, *Panonychus ulmi* (Koch.). Where rates of application approached 2 pounds or more per acre, this species was held in check. At lower dosages schradan had a tendency to induce an increase in the population. Frequently the pest population reached a destructive level. However, natural enemies often developed at a satisfactory rate to take command of the situation before any injury occurred.

Dosages of schradan at 0.75 pound or higher appeared to exert a suppressive influence on the citrus flat mite, *Brevipalpus lewisi* McG. This species is increasing as a pest of walnut.

RESIDUE.—Because of the high oil content of walnut meats, difficulty was encountered in determining the amount of residue present in the harvested nuts. The results obtained are given in table 6. Some residue was encountered in the samples but the amount found did not exceed 0.5 p.p.m., which apparently is a level that would not present a serious health hazard. The residues reported here are in close agreement with the amounts found by Blinn *et al.* (1957). They recovered approximately 0.2 p.p.m. of schradan in harvested nuts from Placentia walnut trees that were sprayed on May 21 with either 1 or 2 pounds of schradan.

DISCUSSION.—Walnut trees in experimental plots treated with schradan have on the whole appeared to be in much better condition than trees in plots treated with other insecticides. The contrast was most pronounced when the schradan treatments were contrasted with those that failed to give adequate control of the walnut aphid. In some cases the difference was so marked that the question arose as to whether all the benefit derived was due entirely to the elimination of the aphid. Although not proven, there is evidence that schradan may exert a stimulating effect beyond the control of pests. Banerjee & Basu (1957) observed a marked increase in the yield of rice in plots treated with schradan. The increase apparently was not associated with the control of an insect pest. Casida *et al.* (1952) noted that at low concentrations schradan accelerated growth, while Zeid & Cutkomp (1951) observed that an increase in the carbohydrate content of plants as well as other changes occurred where plants were treated with the insecticide. The latter workers further noted that the chemical changes in the plants were similar to those produced by the herbicide 2, 4-D and related compounds. Such modifications could conceivably be beneficial and it is possible that this actually occurs in the case of walnuts.

The exceptional ability of schradan to control the walnut aphid has made it possible to evaluate rather accurately the exact damage inflicted on walnuts by the several sucking pests. Where trees have been maintained free of scales and spider mites it has been observed that aphids can reduce the crop by approximately 50%. It has been demonstrated that the most critical period is when severe infestations occur on early varieties from the time the leaf buds start to grow until the leaves are fully expanded. The leaves may be greatly stunted which is re-

Table 6.—Schradan residues found in walnut meats at harvest.

Schradan Applied per Acre (lbs.)	Date Applied	Residue in Walnut Meats (p.p.m.)
1.0	May 2, 1956	0.10
2.0	May 2, 1956	.30
0.5	Apr. 3, 1957	.12
1.0	Apr. 3, 1957	.27
1.0	Apr. 9, 1957	.14
1.0	Apr. 16, 1957	.40
1.0	Apr. 30, 1957	.27

flected in a reduced size of the developing nuts. Also, the destructive influence may be carried over into the second year. Late varieties are usually less subject to aphid damage than the early ones because, with the former, destructive aphid populations seldom develop until after the leaves have expanded.

In the absence of aphids or spider mites it has been determined that severe infestations of soft scales can greatly reduce the vigor and growth of the trees. Trees protected from aphids with schradan, but heavily infested with scales may be of good color but the leaves are much stunted and look less thrifty when compared with those from trees protected from both aphids and scales. The poorer leaf growth is reflected in a lowering of the yield and in reduced twig growth.

The situation that occurs in the case of spider mites is not as sharply defined as that which exists with the walnut aphid and soft scales. The only spider mite that develops in the presence of schradan is the European red mite. Induced increases of this spider mite have resulted in rather serious leaf damage and defoliation. A considerable amount of feeding can occur without greatly lowering the quality of the crop, but a point is reached where weight and color of the walnut meats may be seriously impaired.

Except in the area where the walnut aphid has shown some resistance to phosphate insecticides, 0.5 pound of schradan per acre has resulted in satisfactory control for an entire season. Better results were obtained where the dosage was increased to 0.75 to 1.0 pound per acre. The disadvantage of schradan is its tendency to induce an increase in populations of the European red mite and the calico scale.

Other aphicides tested that show considerable promise are ethion, Trithion and Guthion. These materials appear to be more effective against the aphid than parathion or malathion. Further, all of them appear to be effective against one or more of the other destructive pests of walnut.

ACKNOWLEDGMENTS.—We express our appreciation and thanks to C. C. Anderson, Ralph Bishop, H. E. Minahan, H. S. Hight, Wayne Haas and the several growers for their excellent cooperation in the conduct of these investigations. Thanks are also due the firms which furnished insecticides and other assistance. For field and laboratory help, we wish to thank the following: Fred Charles, Milton Bell, Don Rough, A. T. Gotelli, Donald Burdick and Marius Wasbauer. We extend our thanks for the schradan residue analyses to R. P. Miskus who conducted the study under the supervision of Dr. William Hoskins who is in charge of the residue investigations in the Department of Entomology, University of California, Berkeley, California.

REFERENCES CITED

Banerjee, S. W., and A. C. Basu. 1957. Experiments with systemic insecticides for the control of *Schoenobius incertulas* (Wlk.). (Lepidoptera, Pyralidae), a stem borer of paddy in West Bengal. Bull. Ent. Res. **48** (pt. 2): 299–303.

Blinn, R. C., F. A. Gunther, J. C. Ortega and L. M. Elliott. 1957. Harvest residues of schradan in meats of field-treated walnuts. Jour. Econ. Ent. **50**(4): 500–1.

Casida, J. E., R. K. Chapman and T. C. Allen. 1952. Relation of absorption and metabolism of octamethylpyrophosphoramide by pea plants to available phosphorus. Jour. Econ. Ent. **45**(4): 568–78.

Fjelddalen, J. 1955. Systemic preparations against pests of fruit trees, berry fruits and ornamental plants. Höfchen-Briefe English edition **8**(1): 1–27 Pub. Bayer Plant Prot. Dep. Leverkusen and Höfchen, Germany].

Jefferson, R. N. 1951. Octamethyl pyrophosphoramide and a trialkyl thiophosphate for control of aphids on *Centaurèa cyànus*. Jour. Econ. Ent. **44**(6): 1021–2.

Michelbacher, A. E. 1955. Further observations on the control of the walnut aphid. Jour. Econ. Ent. **48**(5): 504–9.

Michelbacher, A. E., O. H. Fullmer, C. C. Cassil and C. S. Davis. 1954. Walnut aphid resistant to parathion in northern California. Jour. Econ. Ent. **47**(2): 366–7.

Michelbacher, A. E., and Earl Oatman. 1955. Marked suppressing action of schradan on the walnut aphid. Jour. Econ. Ent. **48**(6): 768–9.

Ripper, W. E., R. M. Greenslade and G. S. Hartley. 1950. A new systemic insecticide Bis (Bis dimethylamino phosphonous) anhydride. Bull. Ent. Res. **40**(4): 481–501.

Ripper, W. E., R. M. Greenslade, and G. S. Hartley. 1951. Selective insecticides and biological control. Jour. Econ. Ent. **44**(4): 448–59.

Thomas, W. D. E. 1956. Study with systemics—schradan moves up from the leaves. The Grower, pp. 711 and 713. Grower Publ. Ltd., 49 Doughty St., London.

Zeid, Mahmoud, M. I., and L. K. Cutkomp. 1951. Effects associated with toxicity and plant translocation of three phosphate insecticides. Jour. Econ. Ent. **44**(6): 898–905.

Glycine-Sodium Hydroxide Solution as an Attractant for Walnut Husk Fly[1]

M. M. BARNES and J. C. ORTEGA,[2] *University of California Citrus Experiment Station, Riverside*

ABSTRACT

A solution of 2.25% glycine-3% sodium hydroxide, aged in the absence of walnut husk flies (*Rhagoletis completa* Cresson), rapidly increases in attractive power to this species during the first 2 weeks. From 2 to at least 7 weeks, and probably longer, this solution maintains a steady level of high attractant value. The optimum use of this bait solution in obtaining an index of fly populations in experiments involving area control of adults, and its current use in timing of the first spray for walnut husk fly control, are discussed.

An excellent attractant for the walnut husk fly (*Rhagoletis completa* Cresson) consists of 3 ounces glycine and 4 ounces sodium hydroxide in 1 gallon of water (Boyce & Bartlett 1941). Very few insects other than walnut husk flies are caught in bait pans filled with this solution. Such

[1] Paper No. 1027, University of California Citrus Experiment Station, Riverside, California. Accepted for publication March 10, 1958.

[2] The authors acknowledge the assistance of C. R. Ash in these studies. For the statistical analyses the authors are indebted to M. J. Garber, Assistant Biometrician in the Experiment Station.

bait traps have been useful for timing of spray treatments, since they provide an index to the beginning of fly emergence.

In indexing walnut husk fly abundance for the purpose of studying effects of area treatments upon fly populations, bait traps that maintain a relatively constant power of attraction are desirable. The relative power of attraction of bait solutions of glycine of different ages has previously been studied by Boyce & Bartlett (1941). They found that over a period of 4 weeks, significantly greater catches were obtained when water was added to partially evaporated glycine solutions than when freshly prepared solutions of glycine were substituted for the old material at weekly intervals. This indicates that glycine solutions increase in effectiveness with exposure in the orchard, and that solutions less than 1 week old are less effective than those 1 to 4 weeks old. Similarly, Hodson (1943) reported that a 1% glycine-1% sodium hydroxide solution aged in the orchard for 6 weeks was more attractive to apple maggot flies than solutions aged in the orchard for 2 or 4 weeks.

METHODS AND MATERIALS.—The following experiment was carried out to provide information on the length of time required for a 2.25% glycine-3% sodium hydroxide solution to reach maximum attractive power to walnut husk flies, and the length of time during which the solution maintained attractiveness. The factor of husk-fly contamination was excluded by aging the solutions in the absence of husk flies. Solutions of glycine (Eastman red label) 3 ounces, sodium hydroxide (A.C.S.) 4 ounces per gallon of water were prepared 1, 2, 4, 5, and 7 weeks before placing in the orchard. Each of these solutions was distributed equally among six 1-quart, grey-enameled pudding pans and hung in a lath house for aging in semi-shade and in the absence of a walnut husk fly population. Water was added regularly to replace evaporative loss.

On August 16, at the seventh week, all bait pans, including a freshly prepared series, were hung in trees in a 6×6 Latin-square design in a cling-stone peach orchard infested with walnut husk flies. One pan was hung in every fifth tree in every fourth row in the center area of the 20-acre orchard. The number of flies caught was recorded each day for 6 days. The traps were rotated each day, each trap being exposed for 1 day in each of the "trap trees" used, in the row in which it was originally placed. Daily rotation of traps improves accuracy of results to the extent that it overcomes the effect of location in different trees. This procedure provided data which could be analyzed as six Latin squares, accounting for a great deal of the large amount of variation encountered in such field trials with attractants.

RESULTS.—Results are presented in table 1. From these it will be seen that the fresh solution and the solution 1 week old were considerably less attractive than solutions 2 weeks old or older. The differences are statistically significant. After 2 weeks the solution had reached its peak in attractancy and it maintained this level for a period of at least 7 weeks. Results from bait-pan installations in 1957 suggest a high power of attraction through at least 11 weeks.

The steady level of attraction that this glycine-sodium hydroxide solution maintains for a period of 2 to 7 weeks is a characteristic that is useful in area trials directed against adult flies, where an index of the fly population is desirable for a period of 1 to 2 weeks before treatment and for a period of 2 to 3 weeks after treatment. It follows that installation of pans should be made about 2 weeks before the period of interest.

Table 1.—Effect of aging of glycine-sodium hydroxide solutions on attraction to walnut husk flies.

Age of Solution in Weeks	Total Number of Flies	Average Number of Flies per Trap, per Day[a]	Comparisons[b] at Odds of: 19:1	99:1
0	112	3.1	B C	
1	180	5.0	B C	
2	324	9.0	A	B A
4	271	7.5	A	B A
5	391	10.8	A	A
7	384	10.7	A	A

[a] Average of six replications for a 6-day period, August 16–22, 1956.
[b] By significance tests for ranked treatments. Treatments in same letter group are not significantly different

The fact that the glycine-sodium hydroxide solution increases in attractiveness over a 2-week period has an apparent bearing on its usefulness in timing the first walnut husk fly spray. Grower practice has been to use bait pans as an index of significant emergence and fly activity. Bait pans are placed in the orchard about mid-July. Light and fluctuating catches usually follow for a short period. However, the first spray application is not made until catches become regular and show a steady increase in number. This practice is related to the fact that in many seasons there is trace emergence for an indefinite period, followed by the main emergence. Onset of significant emergence may vary by area and season as much as a month. Apparently, the best use of the glycine-sodium hydroxide bait for detecting significant emergence is to install the bait pans at least 2 weeks ahead of the earliest known period of significant emergence to provide a more reliable index of significant fly activity. This is further demonstrated by results of a comparison between a bait solution 2 weeks old (and catching walnut husk flies during this period), and a freshly prepared solution, both introduced into an orchard with a high fly population. Results from five replicates were as follows:

Solution	*Total Number of Flies Caught* *September 1*	*September 2*
Fresh	34	23
Two weeks old	975	833

It is apparent that the fresh solution provides a catch which is low to the point of being quite misleading as to fly activity. From previous studies on emergence as determined from emergence cages (Boyce 1934) and from observations on earliest onset of significant oviposition which may in some years and in some localities be as early as July 20, and in the light of a 2-week aging requirement, it appears that optimum use of bait pails of glycine-sodium hydroxide solution for timing of the first application would follow by installing these no later than July 1.

Addition of sodium lauryl sulfate increased husk-fly entrapment by the glycine-sodium hydroxide solution

(Hodson 1943) by about 50% as shown from the following 19-day trial with five replications and five rotations:

Solution	Total No. of Flies Caught
A. Glycine 2.25%-sodium hydroxide 3%	433
B. As above with 0.01% sodium lauryl sulfate added	663
C. Sodium lauryl sulfate only, 0.01% in water	53

This increase in catch is significant at odds of 19:1. Ordinarily, the bait solution without addition of a surface active material catches a sufficiently large number of flies to serve its purpose. However, in light populations use of sodium lauryl sulfate may be desirable.

In August 1957, bait pails of glycine-sodium hydroxide solution were exposed in 13 orchards in Los Angeles, Riverside, and San Bernardino Counties. The entire catch from 58 bait pails during the period August 16 to 23, a total of 4,490 flies, was determined as to sex. Of this total, 67.8% were females. The sex ratio in this species is 1:1, but whether the predominant catch of females by the lure is related to a difference in attraction or a difference in entrapment of the sexes has not been determined.

A comparison designed to provide information on the question as to whether husk fly contamination affects catch was not productive of information because of an unexpectedly low population of flies in the trial area.

REFERENCES CITED

Boyce, A. M. 1934. Bionomics of the walnut husk fly, *Rhagoletis completa*. Hilgardia 8(11): 363–579.

Boyce, A. M., and B. R. Bartlett. 1941. Lures for the walnut husk fly. Jour. Econ. Ent. 34(2): 318.

Hodson, A. C. 1943. Lures attractive to the apple maggot. Jour. Econ. Ent. 36(4): 545–8.

The Effect of Insecticidal Soil Drenches on the Growth of Young Cabbage and Muskmelon Plants[1]

WILLIAM G. YENDOL and GEORGE E. GOULD, *Purdue University, Lafayette, Indiana*

ABSTRACT

Soil drenches of insecticidal emulsions are commonly used by commercial growers in Indiana in transplanting cabbage and related cruciferous crops for the control of the cabbage maggot, *Hylemyia brassicae* (Bouché), and occasionally for the control of cutworms around hills of melons. Investigations were made on young cabbage and muskmelon plants grown in the greenhouse to determine the chemicals in the formulations which at certain dilutions have caused retardation of plant growth and even death of plants under field conditions. Emulsion formulations of six insecticides—aldrin, chlordane, dieldrin, endrin, heptachlor and lindane—and emulsion formulations of their respective solvent-emulsifiers were tested at six dilutions on cabbage and 18 dilutions on muskmelon plants. Results based on the number and weight of surviving plants indicated that muskmelon plants were more sensitive to the drenches than cabbage plants and that in general the solvent-emulsifier formulations were more toxic than their respective complete emulsion. Of the six materials tested, formulations of aldrin and chlordane were less toxic to young cabbage and muskmelon plants than formulations of dieldrin, endrin and heptachlor, which in turn were less toxic than the lindane formulations.

Control of soil insects first became possible and practical with the development and release of the chlorinated hydrocarbon insecticides. Many methods of introducing them into the soil have been devised, such as treating seed, mixing with fertilizer, banding in or over the row, broadcasting and working into the soil, and drenching around individual plants or hills. Drenches are being used in Indiana in transplanting cabbage and related cruciferous crops and occasionally for the control of cutworms around hills of melons. Growers, however, have observed that certain insecticidal emulsion formulations caused a retardation of plant growth and even the death of plants. In order to isolate the chemicals in these formulations that might cause this trouble, tests using young cabbage and muskmelon plants were initiated in the greenhouse to compare emulsifiable concentrates of six standard insecticides with the identical formulations without the insecticide.

The drench method of applying emulsions to the soil around the base of plants has given good control of the cabbage maggot, *Hylemyia brassicae* (Bouché), on cabbage and related plants. Under certain soil conditions, however, and with certain materials, stunting, retardation of growth and even the death of young plants resulted (Gould 1955). Growers normally apply about 4 fluid ounces around the plant, either automatically by a transplanting machine or from tanks a few days after the plants are set. Drench applications used by a few melon growers in 1955 to check cutworm damage also caused injury to muskmelons and watermelons. Little information has been published on the phytotoxicity of the various ingredients that go into an emulsion. Wright (1945) found that drenches of a BHC suspension and of chlordane emulsion had no appreciable effect on cauliflower grown in pots in the greenhouse. Stitt *et al.* (1948) used various insecticide emulsions in a starter solution for cabbage and found no injury from chlordane, slight injury from lindane and serious injury from BHC. Eide *et al.* (1950) reported that a chlordane emulsion added to a starter solution caused no injury to cabbage and broccoli transplants,

[1] These data were taken from a thesis submitted by the senior author in partial fulfillment of the requirements for an M.S. degree. Purdue University Agricultural Experiment Station Journal paper No. 1244. Accepted for publication March 17, 1958.

but did have a retarding effect on cauliflower transplanted when temperatures were warm and the soil dry. Gast & Early (1956) investigated the possible toxic effects of emulsifiers in a nutrient solution to tomato and tobacco plants and found that at a 1% concentration only Triton X-45 was injurious, while Atlox 1045A, Triton B-1956 and Triton X-155 were safe.

PROCEDURE.—For these tests cabbage of the variety Marion Market and muskmelons of the variety Pride of Wisconsin were used. Containers for growing the plants were gallon-sized tin cans with holes in the bottom to provide drainage. The cans were partially filled with 6 pounds of a mixture of one part air-dried light silt loam and one part sand. Cabbage plants were started in flats and on the seventh day four were transplanted into each can. Muskmelon seed was planted directly in the cans and on the 10th day the plants were thinned to four. Tests were run in the greenhouse from October to early June. Daytime temperatures during the fall and winter months ranged from 70° to 80° F. and from 75° to 95° in the spring. As a consequence the plants during the winter months were smaller than those grown in May and June when temperatures were higher and light conditions were more nearly optimum. All treatments, including an untreated check with each series, were replicated five times for each dilution for each type of plant.

Emulsifiable concentrates of six insecticides were obtained from the manufacturers. These companies also supplied a mixture of the solvents and emulsifiers (see table 1) identical in proportion and formulation to that in their complete concentrate. The six insecticides used and the supplier of samples were aldrin, dieldrin and endrin from the Shell Chemical Corporation; chlordane and heptachlor from the Velsicol Chemical Corporation; and lindane from the California Spray-Chemical Corporation. The constituents of the emulsifiable concentrates and the percentage of each, except for lindane, are given in table 1. Information of the constituents of lindane were not made available by the manufacturer.

Drenches were applied to the soil around the plants 21 days after the muskmelon seed was planted or 21 days after cabbage was transplanted. The rate of application of drenches by growers is customarily 300 gallons per acre for cabbage and 100 gallons for muskmelons. Since spring cabbage plants are set 9 to 12 inches apart in rows and rows 30 inches apart, the amount of drench to be used per can was determined to be 120 ml. Spacing for muskmelons in the field is normally 5 by 5 feet and thus at 100 gallons per acre the equivalent amount used in the greenhouse was 210 ml. per can. Stock emulsions for a 1:99 dilution were prepared by adding a sufficient quantity of the emulsifiable concentrate to give one part of the actual insecticide in 99 parts of the combined solvent, emulsifier and water. To avoid confusion each solvent-emulsifier mixture was diluted in the same proportion as the complete emulsifiable concentrate. In tables 2 and 3 results of treatments with solvent-emulsifier and the solvent-emulsifier plus insecticide may be compared, since the complete series of both were applied the same day.

The standard dilutions used on cabbage were 1:99, 1:199, 1:399, 1:799, 1:1599 and 1:3199. On muskmelons a first series of tests was made at these dilutions, but later a second series with more dilutions was run to determine with greater accuracy the safe concentrations. Five days after each treatment plants were removed from the soil and weighed and leaves and roots examined for possible injury.

RESULTS WITH CABBAGE.—Drenches prepared from aldrin, chlordane and dieldrin formulations caused little

Table 1.—The percentage by weight of the technical and actual insecticide, solvents and emulsifiers in six commercial emulsifiable concentrates and in identical formulations with only the solvents and emulsifiers.

	ALDRIN		CHLORDANE[a]		DIELDRIN[a]		ENDRIN[a]		HEPTACHLOR[a]		LINDANE[a]	
	Per Cent		Per Cent		Per Cent		Per Cent		Per Cent		Per Cent	
MATERIALS	A	B	A	B	A	B	A	B	A	B	A	B
Insecticide												
Technical	28.1	0	75.0	0	19.0	0	21.6	0	33.5	0	—	0
Actual	25.3	0	72.0	0	19.0	0	19.7	0	24.1	0	20.0	0
Solvents												
Ethyl acetate									43.5	65.4		
Isopropanol									13.0	19.6		
Kerosene	32.2	44.8	20.0	80.0								
Shell TS-28R	32.2	44.8										
Xylene					75.0	92.6	70.9	90.4				
Emulsifiers												
Atlox 1045A	3.0	4.2										
Atlox G1690									10.0	15.0		
Emcol H83T			5.0	20.0								
Epichlorohydrin	0.5	0.6										
Trem Y-124	1.0	1.4										
Triton X-45	3.0	4.2										
Triton X-155					4.0	4.9	5.0	6.4				
Triton B-1956					2.0	2.5	2.5	3.2				
Other ingredients											80.0	100.0
Toxicant per gal. (lb.)	2.0	0	8.0	0	1.5	0	1.6	0	2.0	0	1.6	0

[a] A, complete insecidide; B, solvents and emulsifiers only.

Table 2.—The survival and weight of cabbage plants treated with drenches containing emulsifiable concentrates and identical formulations with only the solvents and emulsifiers.

Dilutions	Aldrin[a]		Chlordane[a]		Dieldrin[a]		Endrin[a]		Heptachlor[a]		Lindane[a]	
	A	B	A	B	A	B	A	B	A	B	A	B
	Plants surviving (per cent)											
1:99	100	100	100	100	100	80	15	5	5	20	100	100
1:199	100	100	100	100	100	80	15	0	20	20	100	100
1:399	100	100	100	100	100	100	65	32	31	57	80	15
1:799	100	100	100	100	100	100	100	70	100	90	95	80
1:1599	100	100	100	100	100	100	95	95	93	93	100	100
1:3199	100	100	100	100	100	100	100	100	100	100	100	100
Untreated	100	100	100	100	100	100	100	100	100	100	100	100
L.S.D. at 5% level	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	22	22	31	31	n.s.	17
at 1% level	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	32	30	43	43	n.s.	23
	Weight of plants (grams)											
1:99	0.39	0.45	1.71	1.89	2.51	1.67	0.34	0.11	0.45	0.37	2.11	2.05
1:199	0.40	0.46	2.47	1.80	2.42	1.53	0.26	0	0.29	0.31	1.70	1.95
1:399	0.38	0.45	2.05	1.65	1.90	2.69	0.57	0.21	0.96	1.16	1.32	1.00
1:799	0.46	0.71	1.94	1.59	3.66	2.42	0.59	0.65	1.50	1.41	1.86	1.98
1:1599	0.45	0.52	1.85	1.24	4.33	3.11	1.09	0.96	1.75	1.30	2.37	1.99
1:3199	0.78	0.71	2.76	1.28	5.22	3.59	0.86	1.14	1.87	1.15	2.75	2.51
Untreated	0.54	0.54	1.88	1.88	5.08	5.08	1.38	1.38	2.27	2.27	2.91	2.91
L.S.D. at 5% level	n.s.	0.16	n.s.	n.s.	1.56	n.s.	0.39	0.39	0.79	0.57	0.68	0.78
at 1% level	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	0.53	0.53	1.07	0.77	n.s.	n.s.

[a] A, complete insecticide; B, solvents and emulsifiers only.

harm to young cabbage plants, as all plants, with few exceptions, survived the treatment. The weights of surviving plants were uniform in most instances (table 2). Both endrin and heptachlor formulations at dilutions of 1:99, 1:199 and 1:399 had some deleterious effect, as they caused the death of numerous plants and reduced the average weight of survivors. Severity of plant injury was about equal for the solvent-emulsifier and the solvent-emulsifier plus insecticide. Of the two insecticides the endrin formulations were more toxic. Results with the lindane formulations were inconclusive, as no injury occurred at the lowest and at the highest dilutions, but some did occur in the middle range.

Results with Muskmelons.—Young muskmelon plants were found to be more sensitive to the test formulations than were the young cabbage plants. Series of tests run at 1:99, 1:199, 1:399, 1:799, 1:1599 and 1:3199 were not conclusive as to the dilution range at which injury would occur and so a second series of tests were run at many more dilutions intermediate to the original six. The results are given in table 3. A difficulty in interpreting results arose from the different concentrations of the insecticide, ranging from 1½ pounds of actual toxicant per gallon for endrin and dieldrin up to 8 pounds for chlordane. Consequently it was necessary to use some materials, such as chlordane, at dilutions of 1:99 and lower.

Survival of plants and their average weight indicated that for chlordane the solvent-emulsifier and the same with the insecticide added were about equal in toxicity, as both caused a high mortality at a 1:24 dilution and some at 1:49. Both aldrin formulations caused the death of melon plants at a 1:199 dilution and some loss even at 1:399. Both the dieldrin and endrin formulations were more injurious than either aldrin or chlordane. The complete dieldrin and endrin formulations killed all plants at a 1:399 dilution, while their solvent-emulsifiers killed most plants at a 1:199 dilution. A safe concentration for the complete dieldrin emulsion was around 1:1499 and for its solvent-emulsifier at 1:2799; for the complete endrin emulsion at around 1:1399 and for its solvent-emulsifier at 1:2399. The heptachlor formulations were not as phytotoxic as those of dieldrin or endrin, but again the solvent-emulsifier was more toxic than the complete formulation. Lindane was one of the more injurious formulations, as the safe dilution for the complete emulsion was 1:1799 and for its solvent-emulsifier around 1:1399.

The data on weight of plants was in general agreement with the figures for survival. This was especially true for aldrin, chlordane and dieldrin, although differences did occur in the other three which were grown in the spring. The weight of untreated plants was significantly greater than even the weakest dilutions of both heptachlor formulations, both lindane formulations and the complete endrin. In general plants treated with the complete emulsions grew better and were larger than those treated with their respective solvent-emulsifiers alone.

Discussion.—The effect of any one solvent or emulsifier on the phytotoxicity of its respective formulation

Table 3.—The survival and weight of muskmelon plants treated with drenches containing emulsifiable concentrates and identical formulations with only the solvents and emulsifiers.

Dilutions	Aldrin[a]		Chlordane[a]		Dieldrin[a]		Endrin[a]		Heptachlor[a]		Lindane[a]	
	A	B	A	B	A	B	A	B	A	B	A	B
					Plants surviving (per cent)							
1:24			45	40								
1:49			63	95								
1:74			80	100								
1:99	0	0	100	100	0	0	0	0	0	0	0	0
1:199	3	0	95	100	0	0	0	0	0	0		
1:299	30		100	100								
1:399	70	26	100	100	0	0	0	0	0	0	0	0
1:499	90	100							45			
1:699	100	100							82	60		
1:799					15	0	25	0	100	75	5	0
1:899					6	0	25	0	80	95	15	
1:999					32	0	45	5	100	86	40	0
1:1099					38		46			81		66
1:1199					61	20	70	35	100	100	30	60
1:1399					45	50	95	26	100	100	33	75
1:1599					85	65	100	40		100	50	100
1:1799					100	62		60			80	100
1:2399					100	61		85			100	
1:2799						100		100				
Untreated	100	100	100	100	100	100	100	100	100	100	100	100
L.S.D. at 5% level	30	28	21	n.s.	36	42	33	43	26	24	35	43
at 1% level	40	38	28	n.s.	49	56	44	n.s.	n.s.	n.s.	46	58
					Weight of plants (grams)							
1:24			1.65	0.71								
1:49			1.97	2.10								
1:74			2.89	2.30								
1:99			2.67	2.80								
1:199			2.71	2.84								
1:299	0.92		2.47	2.85								
1:399	1.44	1.61	2.44	2.86								
1:499	1.77	3.16	2.67	2.86					2.69			
1:699	1.83	2.73							3.39	2.63		
1:799					0.50		1.09		4.57	4.12	0.75	
1:899					0.34		2.23		4.95	4.19	0.80	
1:999					0.72		1.84	0.39	4.99	3.94		
1:1099					0.69		3.00			4.15	2.67	4.60
1:1199					1.01	0.60	3.44	0.76		5.33	2.40	2.91
1:1399					0.96	2.65	3.65	2.15	6.06	5.95	1.89	4.28
1:1599					1.25	2.84	3.92	2.22		5.74	3.92	5.84
1:1799					1.38	2.27		2.02			5.53	5.73
1:2399						2.69		3.05				
1:2799						3.00		3.10				
Untreated	2.36	3.77	3.67	2.64	2.05	4.90	6.66	4.53	8.78	9.19	8.01	8.40
L.S.D. at 5% level	0.42	1.19	n.s.	0.92	0.71	1.18	1.74	1.54	1.65	1.53	1.73	2.84
at 1% level	0.57	1.60	n.s.	1.23	n.s.	1.58	2.33	2.06	2.22	2.05	2.32	3.81

[a] A, complete insecticide; B, solvents and emulsifiers only.

has been difficult to evaluate, as such small quantities were present in the finished emulsions. For example, xylene made up 75% of the dieldrin concentrate and 71% of the endrin and yet at the strongest dilution, 1% of the actual toxicant, it made up only 4.7 grams of the former and 3.5 grams of the latter in 95 and 94 grams of water. The two solvents for aldrin, kerosene and Shell TS-28R, made up 64% of the concentrate, but the two aldrin emulsions caused less injury than dieldrin and endrin. Heptachlor with 56.5% solvents caused more injury than aldrin. The quantity of emulsifiers present was small, the largest being in the 1% heptachlor, where 0.41 grams were used with 3.73 grams of toxicant and solvent and 95.86 grams of water.

From these tests it can be concluded that the complete formulations of the six emulsifiable concentrates were as safe as, or slightly safer than, formulations prepared from the solvent-emulsifier alone; and that the formulations of aldrin and chlordane were less toxic to young cabbage and muskmelon plants than were the formulations of dieldrin, endrin and heptachlor, which in turn were less toxic than the lindane formulation.

REFERENCES CITED

Eide, Paul M., and Loyd L. Stitt. 1950. Comparison of insecticides for cabbage maggot control. Jour. Econ. Ent. **43**(6): 899–905.

Gast, Robert, and Jack Early. 1956. The phytotoxicity of solvents and emulsifiers used in insecticidal formulations. Agric. Chemicals **11**(4): 42–45, 136, 137, 139.

Gould, George E. 1955. Cabbage maggot control. Purdue University Agri. Expt. Sta. Bull. **616.**

Stitt, Loyd L., and Paul M. Eide. 1948. New insecticides for cabbage maggot control in Western Washington. Jour. Econ. Ent. **41**(6): 865–9.

Wright, D. W. 1954. The control of cabbage root fly on cauliflower plants raised in pots. 4th Ann. Rept. (1953), Natl. Veg. Res. Sta., Wellesbourne, Warwick, England, pp. 21–26.

BOOK REVIEW

A CRITICAL REVIEW OF THE TECHNIQUES FOR TESTING INSECTICIDES, by J. R. BUSVINE. 1957. Published by Commonwealth Institute of Entomology, 56 Queen's Gate, S. W. 7, London, England, 208 pp.

This book should be extremely valuable to research workers engaged in the study of insecticides, for it contains descriptions of many of the popular testing techniques that have been used in recent years. The author has done an excellent job of covering the literature and has presented his information in an interesting and readable form. The task of assembling it must have been a monumental one for he used 551 manuscripts published between 1909 and 1957 as references. Dr. Busvine believes too many entomologists insist on developing their own testing techniques instead of seeking uniformity. Publication of the book may help reverse such a tendency by making the methods that already have been developed available to the many investigators who do not have access to adequate library facilities.

Included in the 10 chapters are descriptions of methods that have been used to test stomach poisons, contact poisons, fumigants, and repellents. They are applicable for use against insects that attack agricultural crops, stored products, man, and animals. There are also chapters on apparatus for conducting laboratory tests by immersing, spraying, or injecting the insects, and on methods of analyzing data statistically.

In addition to descriptions of testing techniques, the physical, mechanical, and chemical factors that affect insecticides and repellents are discussed fully. Chapter 3 deals exclusively with this type of information, even though it is titled "Standardization of Insects for Testing." No suggestions are given for obtaining uniform test insects, but convincing evidence is presented from the literature to show that specificity of action, environmental conditions, as well as sex, age, and size of the insects may have an effect on the results obtained with insecticides and repellents. In other chapters such items as (1) particle size, (2) factors affecting suspensions, emulsions, or solutions of insecticides, (3) characteristics of aerosols, and (4) physical factors involved in making topical applications or injections are covered under appropriate chapter headings.

After a critical reading the reviewer has only two minor complaints to offer. Some confusion may result from the discussion of spray nozzles that produce droplets ranging from 100 to 500 microns in diameter under the heading of aerosols after this term has been defined as particles in the 0.1–50 micron range. Included under aerosols are some of the testing methods that are commonly called space spray techniques in this country. The author also reports the earliest residual insecticidal treatments made with contact insecticides probably were films of non-volatile white oil containing pyrethrins which were used against pests of stored food in warehouses. A report of this work was published by Potter in 1938. Without making a search of the literature the reviewer recalls that beta naphthol was used as a contact insecticide to control overwintering codling moth larvae as early as 1926–27. It was applied on corrugated paper bands that were wrapped around the trunks of apple trees. Most of the stomach poisons that have been used against leaf-feeding insects have depended on their residues to produce a portion of the control, so this type of treatment is not a recent development.

The book is well printed on good quality paper and is attractively bound in a cloth cover. It should prove a welcome and popular addition to entomological literature. The reviewer wishes to compliment the author on his achievement.

JAMES B. GAHAN

SCIENTIFIC NOTES

Injury of Dairy Cattle by Sodium Arsenite Spray[1]

A. C. Miller, *Gulf Research and Development Company, Pittsburgh, Pa.*

On July 7, 1956, severe skin injury, mistakenly attributed to an oil-base cattle spray, occurred to a herd of 25 grade dairy cows at a farm near Pittsburgh, Pennsylvania. Observations were made on the injured animals over a period of 5 months. Examination of the cows by one veterinarian 4 days after injury was noted and by another veterinarian 10 days after the injury was first seen revealed no explanation for the severe injury other than that possibly it was being caused by applications of an oil-base cattle spray. Microscopic examinations of the affected areas by veterinarians revealed no parasites or fungi.

The cattle spray which was being used by the dairyman is a conventional oil-base formula containing 0.07% pyrethrins and 0.19% piperonyl butoxide in a highly refined petroleum base oil (typical viscosity of 39.1 SUS at 100° F. and a typical unsulfonated residue of 95.5%). Many hundreds of thousands of gallons of the spray had been used satisfactorily over a period of many years.

The injured areas on the backs and sides of the dairy cows were as much as 1 to 2 square feet in area and were characterized by thickening and hardening of the skin and later by sloughing of large patches of epidermis, with subsequent infection by blow flies in a few cases. Injury was so severe that two of the cows were sent to slaughter at a loss shortly after the condition was first noted, and later three more cows were sold at a loss. Within a few days after injury milk production for the herd dropped 40%. Figure 1 shows typical injuries 27 days after they were first noted.

The location of the injured areas on the backs, sides, and hind quarters of the cows followed the pattern of spray distribution demonstrated by the dairyman who was experienced in the use of oil-base livestock sprays. The hand sprayer used in applying the spray was tested and judged to be suitable for this purpose. It was a new sprayer, and, according to reports, had never contained anything other than the previously described livestock spray. Trials of the sprayer showed that it would have been physically impossible to apply sufficient oil-base spray to cause the injury suffered by the cattle. Several dry cows and beef cattle pastured and housed with the dairy herd, which had not been sprayed, showed no signs of injury.

Inspection of the barn showed several "empty" and partially filled cans of the cattle spray. Spectrographic analysis of small samples of spray from the "empty" and partially filled cans showed that the product was normal.

A more thorough search of the barn revealed an old can of fly spray (not cattle spray). According to the lithographed label on the can, the spray was a conventional oil-base formula designed for use as a space spray against flying insects. Faintly scratched on the lithographed label was the notation "Weed Spray." Examination showed that the can contained an aqueous solution. Chemical analysis of the solution showed it contained 60% by weight sodium arsenite (weed killer concentrate).

Although the sprayer had been filled several times with oil-base cattle spray, the interior and exterior of the sprayer were carefully rinsed with distilled water. Analysis of the residues from these washings showed strong chemical tests for sodium and arsenic. Tests of scab tissue, taken about 6 weeks after injury was first noted, showed that the ash contained about 100 p.p.m. arsenic.

Arrangements were made with the veterinary research department of a state university to conduct tests with the sodium arsenite solution and the oil-base cattle spray. Two Holstein calves, weighing approximately 340 to 400 pounds, were used in these tests. On each animal an area 12 inches square was marked off on the shoulder and flank. Dosages of 1.5 ml. and 3.0 ml. of the two test sprays were applied to these areas at 6.5 p.s.i. gauge pressure with an air brush. Seven applications were made over a period of 3.5 days. One calf received the sodium arsenite spray and the other the oil-base cattle spray. After four applications, a severe thickening and hardening of the skin was detected on the area receiving the 3.0-ml. dosage of sodium arsenite spray. Six days later this area showed severe hardening and marked swelling which extended beyond the sprayed area. Fifteen days after the first application, the area receiving the 1.5-ml. dosage of sodium arsenite had begun to crack and peel, and in 2 months it was almost completely healed. After 2 months, the area receiving the 3.0-ml. dosage, although reduced to 75% of the area sprayed, was still hard, necrotic, and sloughing. Areas treated with the 1.5 and 3.0 ml. dosages of the oil-base cattle spray were normal. The injuries from the sodium arsenite spray appeared identical to those suffered by the dairy cows.

Based on these findings, it was concluded by the spray manufacturer, the veterinarians, and the dairyman that in some unexplained manner the sodium arsenite solution was accidentally applied to the dairy herd. It seems likely that a total of about 1 pint of the spray was applied to the herd over two or three sprayings.

Several large manufacturers of oil-base cattle sprays were asked if they knew of any similar cases of injury. All reported

[1] Accepted for publication June 9, 1958. Partial cost of publication of this paper was met by the Gulf Research & Development Company, Pittsburgh, Pa.

Fig. 1.—Typical injuries to a dairy cow 27 days after the injuries caused by sodium arsenite spray were first noted.

cases of injury caused by gross over-application of normal oil-base products, but none could report a clear-cut case of injury caused by accidental use of a material not designed as a cattle spray.

This case illustrates how a normal product can be mistakenly cited as a cause of injury, and shows the necessity for proper labeling of re-used containers. It also shows that highly toxic materials, such as sodium arsenite, should be stored well-removed from frequently used and much less toxic materials, such as fly and cattle sprays.

The case was considered closed 19 weeks after the injury was first noted. At that time a number of the cows still showed typical injuries, although the scabs were generally much reduced in size.

The Toxicity of Sevin to Goldfish[1]

H. L. HAYNES,[2] H. H. MOOREFIELD, A. J. BORASH, and J. W. KEAYS, *Boyce Thompson Institute for Plant Research, Inc., Yonkers, New York*

Sevin,[3] (1-naphthyl *N*-methylcarbamate), a compound which has displayed broad insecticidal properties (Haynes *et al.* 1957), is currently being extensively evaluated and developed for commercial use. Laboratory and field tests have shown that the gypsy moth, eastern tent caterpillar, sawflies, sawyers, bark and ambrosia beetles are among the insects that may be controlled by this new insecticide. This potential utility against forest insects has prompted determination of the effects of Sevin on wildlife. A preliminary study on the toxicity of Sevin to the common goldfish (*Carassius auratus* (L.)) is presented.

METHOD.—Each test consisted of five goldfish of uniform size (2.5 to 3 inches long), contained in battery jars with 2000 ml. of tap water. Desired concentrations of the technical compounds were dissolved in 1 ml. of ethanol which was added to the water containing the fish at the start of the experiment. The wettable powder formulation (50% Sevin) was weighed out in terms of active ingredient, and stirred directly into the water. Control jars received equivalent quantities of ethanol or wettable powder ingredients minus the toxicant. From four to six replications were made at each concentration in the dosage series, and the tests were repeated on a second batch of goldfish.

During the 48-hour experiments, the fish were held without feeding, the animals were removed immediately after death, and the final mortality counts were made at the end of this time.

Table 1.—Comparative toxicity of insecticides to goldfish.

TREATMENT	INSECTICIDE (P.P.M.)	AVERAGE PER CENT MORTALITY	LD_{50} (P.P.M.)
DDT, technical	0.5	100	0.11
	0.25	90	
	0.125	40	
	0.063	35	
Sevin, technical	35.0	100	28
	30.0	85	
	25.0	20	
	15.0	20	
Sevin, 50%, W.P.	25.0	100	14
	20.0	75	
	15.0	55	
	7.5	0	
Control (ethanol)	0	0	
Control (W.P. ingredients)	0	0	

Average results of all observations are given. The ambient temperature varied from 68° to 78°F.

DISCUSSION.—The data presented in table 1, relative to the toxicity of the reference compound (DDT) are in agreement with observations of other investigators (Ginsburg 1949). By comparison, the goldfish were capable of tolerating over 200 times as much technical Sevin as technical DDT. Under conditions of this test, the wettable powder formulation appeared to be twice as toxic as the Sevin alone. The increased toxicity may be a function of solubility of the insecticide as no mortality was experienced in the control containing only the inert constituents of the powder.

Although Sevin appears to be relatively harmless to goldfish in controlled laboratory tests, under field environment different degrees of toxicity to game or commercial fish may be encountered as a result of variations in formulation, species tolerance or modified feeding habits. Under certain conditions the hazard may be decreased by the instability of Sevin which is susceptible to degradation by alkaline hydrolysis.

REFERENCES CITED

Ginsburg, Joseph M. 1949. Toxicological tests with two new insecticides on mosquitoes and fish. Proc. Ann. Meet., New Jersey Mosquito Extermin. Assoc. **36:** 88–91.

Haynes, Harry L., Joseph A. Lambrech, and Herbert H. Moorefield. 1957. Insecticidal properties and characteristics of 1-naphthyl *N*-methylcarbamate. Contr. Boyce Thompson Inst. **18:** 507–13.

[1] This investigation was sponsored by a Union Carbide Chemicals Co. Fellowship. Accepted for publication May 19, 1958. Partial cost of publication of this paper was met by Union Carbide Chemicals Co., New York, N. Y.

[2] Now with National Carbon Co., New York City.

[3] Sevin is the registered trademark of Union Carbide Corporation.

A Method for Field Studies of the Balsam Woolly Aphid[1]

NORMAN E. JOHNSON and H. J. HEIKKENEN, *Weyerhaeuser Timber Company, Forestry Research Center, Centralia, Washington*

The balsam woolly aphid (*Chermes piceae* Ratz.) is causing serious mortality of Pacific silver fir (*Abies amabilis* (Dougl. Forbes) in the Pacific Northwest. In this region the insect is of recent introduction (Johnson & Wright 1957), but it has been studied intensively in eastern North America by Balch (1952) and in its native Europe (Varty 1956). Marked dissimilarities of environment have necessitated the initiation of studies on the life history of the balsam woolly aphid in the Pacific Northwest. These studies have been facilitated by a micrographic technique devised for identifying and re-examining individual insects *in situ* in bole infestations on mature trees. The present note describes the essential elements and how the technique is used.

Karafiat (1955) has, in studying population dynamics of the balsam woolly aphid in Europe, used a special binocular microscope mounted on a metal staff. This microscope, capable of lateral and horizontal movement, was used to traverse a 3×5 cm. study plot to locate each insect. Karafiat & Franz (1956) used this same microscope, but plotted the position of each insect by means of a pantograph secured to the tree.

The present micrographic technique essentially consists of an assembly of a dissecting microscope with mountings for securing it to the tree and a net reticule placed in one eyepiece to provide grid coordinates. A flashlight is fastened to the body of the microscope to illuminate the insects on the bark, since daylight is rarely adequate under the dense true-fir mixed forest on the west slope of the Cascades. The mountings for the microscope include a slotted bar secured with bolts and wingnuts across

[1] Accepted for publication March 17, 1958.

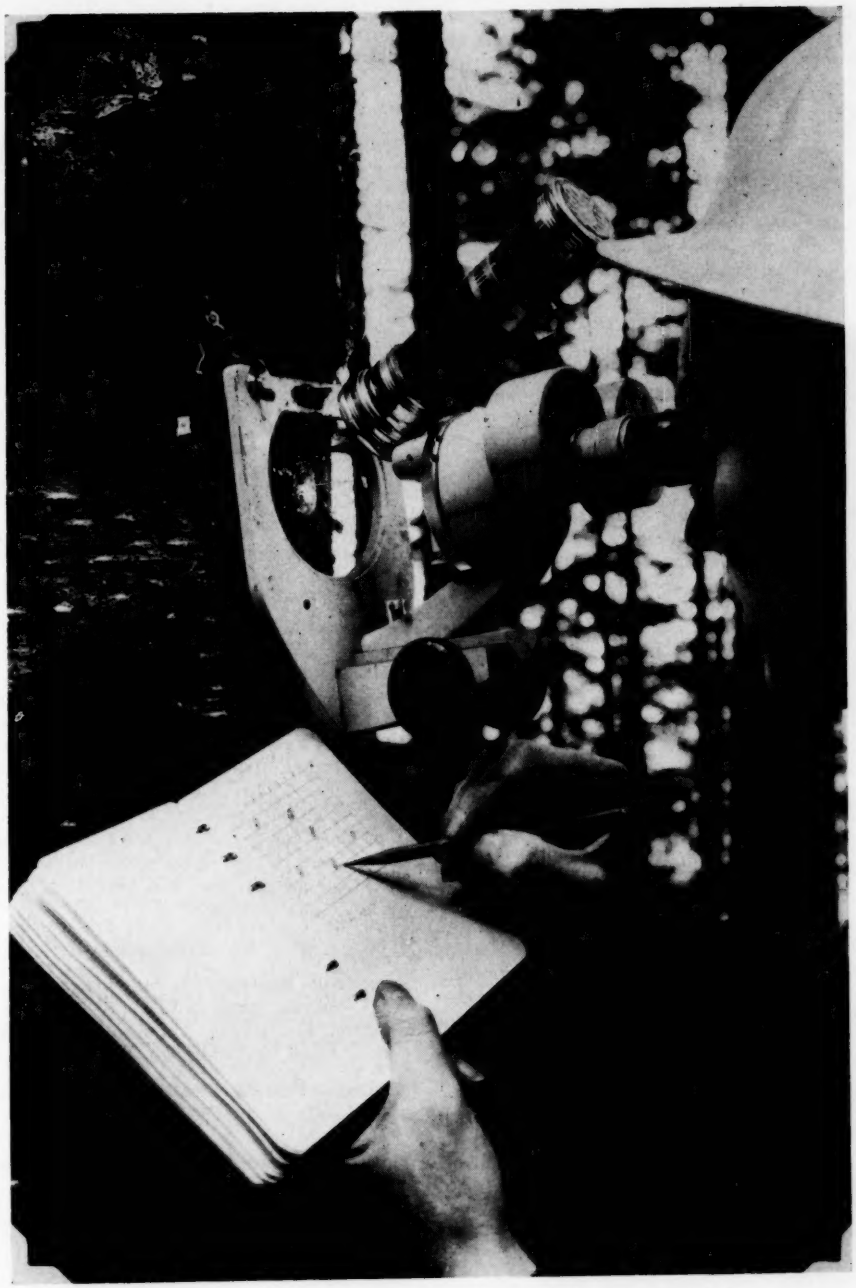


FIG. 1.—Microscope and grid system for following the life history of the balsam woolly aphid *in situ*.

the open end of the base, and a bolt sharpened on one end and screwed into the tapped hole used in securing the microscope to the carrying case. These mountings can be removed easily in order to release the instrument for other uses. The net reticule is a 10×10 mm. square subdivided by 10, numbered, vertical and horizontal lines spaced at 1-mm. intervals. The flashlight is held by a screw-type adjustable hose clamp fitted with a long bolt bent at the proper angle to seat in the illuminator hole found on most dissecting microscopes.

For each study plot location, a headless, sharpened stove bolt is driven into the tree on which the slot on the crossbar is seated, and then the microscope is secured by a wing nut. The sharpened bolt screwed in the base of the microscope acts as a spur in the bark to prevent shifting of the instrument. In order to insure exact relocation of the grid an insect-mounting pin, bearing a waterproof tag with the plot number, is placed in the bark at one corner of the plot. The position of each insect accurately located by means of the grid coordinates of the net reticule is plotted on a form representing an enlargement of the study plot. By periodic re-examination and recording of the insect's stage of development, it is possible to trace the life history and investigate the biology of the balsam woolly aphid. This is facilitated by a population consisting only of wingless sistens that remain stationary after inserting their stylets into the bark. On a plot

size of 0.5 cm. square given by using a 9× eyepiece and 2× objective, over 100 first instar larvae have been mapped and 200 would be possible without stereotyping. The size of the plot may be varied by a change of the magnification or size of reticule.

The grid system mounted in a standard dissecting microscope modified for field use has been a valuable tool in studying the life history and habits of the balsam woolly aphid in the Pacific Northwest. This technique provides the necessary magnification and means of relocating specific insects for periodic study. The adaptations are simple and do not limit the microscope from other uses; it is easily transported, readily and accurately set up, and can be operated by one man.

References Cited

Balch, R. E. 1952. Studies of the balsam woolly aphid *Adelges piceae* Ratz. and its effects on balsam fir, *Abies balsamea* L. Mill. Canada Dept. Agric. Pub. **867.**

Johnson, N. E., and K. H. Wright. 1957. The balsam woolly aphid problem in Oregon and Washington. Pacific Northwest Forest and Range Expt. Sta. Res. Note **18:** 1–34.

Karafiat, H. 1955. Neue Wege der Populationsanalyse an rindenbewohnenden Arthropoden. Nachrichtenblatt des Deutschen Pflanzenschutzdienstes. Jahrgang **7**(8): 133–6.

Karafiat, H. and J. Franz. 1956. Studien zur Populationdynamik der Tannenstammlaus *Adelges* (*Dreyfusia piceae* Ratz.) (Hemipt., Adelgidae) Zool. Jahrb. **84**(6): 467–504.

Varty, I. W. 1956. Adelges insects of silver firs. Forest Comm. [Gt. Brit.] Bull. **26.**

Tests on Control of the Codling Moth in Maine During 1957[1]

L. W. Boulanger, *Maine Agricultural Experiment Station, Orono*

As observed by Siegler & Simanton (1915), the codling moth (*Carpocapsa pomonella* (L.)) seasonal cycle in Maine consists primarily of a single generation with the second generation accounting for only 1 to 2% of the first brood larvae entering overwintering quarters. Bait trap studies in 1957 substantiated this to some extent; of 387 moths captured during the season, only 10 or slightly over 2% were of the second generation.

Control is thus made relatively easier and although codling moth injury is ordinarily kept at a minimum in Maine orchards by the use of lead arsenate alone, it does become necessary on occasion to supplement this program with other materials as indicated by Lathrop (1955). The accepted practice has been to reduce the amount of lead arsenate from 3 to 2 pounds and to add 2 pounds of 50% DDT wettable powder to the 100 gallons of spray mixture. Results obtained with this combination have been very good in the past and although no resistance problems are expected for some time, it was believed that a comparison of the above compounds with other accepted materials, as well as with experimental materials would be in order.

The site selected for the trial was a mixed variety orchard in Cumberland County where codling moth injury during 1956 had been estimated to be over 50%. Single tree plots were replicated 3 times all in 2 rows of McIntosh trees.

Recovery of overwintering larvae before moth flight indicated that a heavy population was present. Bait traps were hung and maintained in another section of the orchard to recover information on moth activity; substantial captures were made throughout the season.

The plots were sprayed on June 4, 17, July, 2, 13, 25, and August 6, the calyx, 1st, 2nd, 3rd, 4th, and 5th cover sprays of that season. The application made on August 6 was incomplete; equipment failure occurred as the last tree in plot 5 was being sprayed. Repairs could not be made until 1 week later and it was believed that spraying the remainder of the plots at this late date would have been of little value.

Table 1.—Results of insecticide tests for codling moth control. Sabbathday Lake, Maine. 1957.

Plot No.	Material and Lbs./100 Gal.		Injury per 100 Fruit[a]	
			Worms	Stings
1	Lead arsenate	3	5.0	10.6
2	Lead+Malathion 25W	2+2	1.8	9.2
3	Lead+DDT 50W	2+2	4.9	8.9
4	DDT 50W	2	4.1	7.4
5	Methoxychlor 50W	2	2.8	3.2
6	DDD 50W	2	19.8	14.8
7	Trithion 25W	2	2.1	4.2
8	Sevin 50W	2	1.0	3.2
9	Ethion 25W	2	6.2	7.2
10	Perthane 50W	2	26.6	7.7
11	Check		50.0	6.8

[a] Average of three replicates.

Applications were made with a dilute hydraulic machine and a hand gun operated from the ground. Trees were sprayed to the run-off stage. Fungicides were applied by the grower in routine sprays.

The data in table 1 show that the most effective control was obtained with Sevin (N-methyl-1-naphthylcarbamate) and the lead arsenate-malathion combination. Methoxychlor and Trithion also gave very good results but the remaining materials particularly Perthane and TDE (DDD) permitted rather heavy larval injury. Lead arsenate alone did not give adequate control as expected but the failure of DDT to give better control when used alone and in combination with lead arsenate is not clearly understood. In view of the rather limited use of DDT in Maine up to the present time, it is very doubtful that any resistance to DDT has developed.

References Cited

Siegler, E. H., and F. L. Simanton. 1915. Life history of the codling moth in Maine. U. S. Dept. Agric. Bull. **252.**

Lathrop, F. H. 1955. Apple insects in Maine. Maine Agric. Expt. Sta. Bull. **540:** 17–20.

[1] Accepted for publication March 10, 1958.

Carabid Beetles Damage Douglas-Fir Seed[1]

James Dick and Norman E. Johnson, *Weyerhaeuser Timber Company, Forestry Research Center, Centralia, Washington*

Damage to Douglas-fir seed different from that caused by mice or shrews was noted during an evaluation of rodent activity on recent cutovers in western Washington. Damage occurred under bark or wood chips placed over seed to protect it from rodents. Carabid beetles found with the damaged seed were suspected of being the causal agent. Specimens brought into the laboratory were presented Douglas-fir seed and alternative vegetable foods. The beetles readily fed on the seed and other materials. Damage to Douglas-fir seed was unmistakably the same as that observed in the field. Most carabid beetles are predators of other insects and certain small animals. However, Essig (1942) reports that certain genera feed on berries, seeds, tender shoots and pollen and foliage of plants. Imms (1948) reports that species of *Harpalus*, *Zabrus*, *Omophron* and *Amara* sometimes devour cereals and seeds of plants. Carabid beetle

[1] Accepted for publication March 17, 1958.

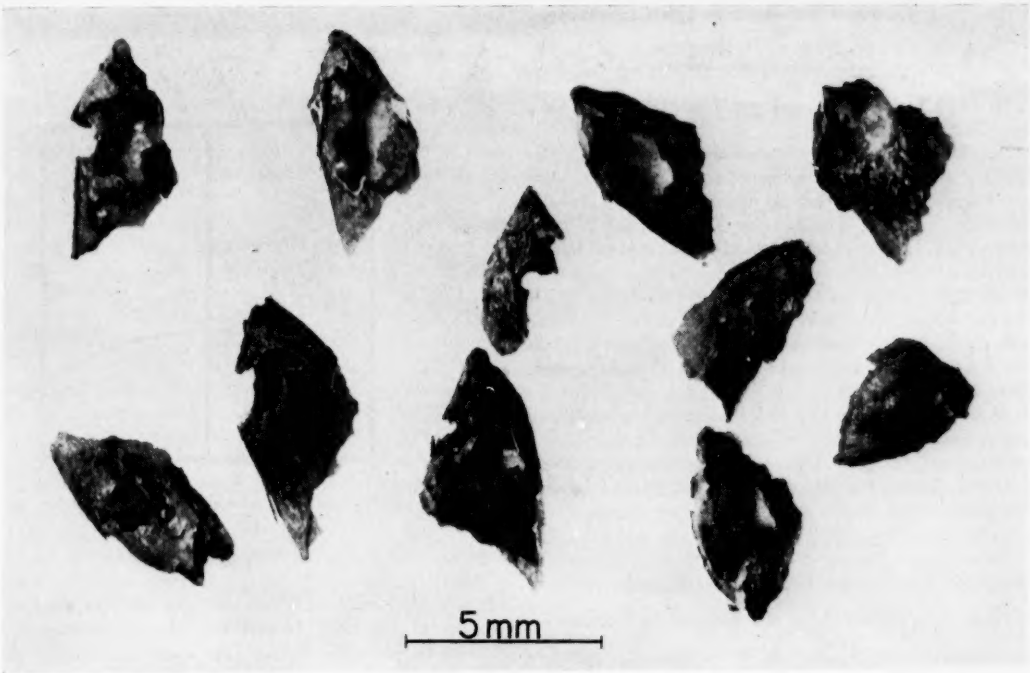


FIG. 1.—Douglas-fir seed damage by *Harpalus cautus* Dejean. Note ragged edges.

damage to coniferous seed in Europe is common. Nüsslin & Rhumbler (1922) indicate that *Harpalus pubescens* Müll. feeds on the seed and seedlings of conifers. These authors point out that other carabid beetles of the genera *Pterostichus* and *Calathus* may feed on seed in nurseries. Nitzsche (1893) reports that carabid beetles of the genus *Harpalus* destroyed up to 80% of the seed and seedlings in nursery beds. Gäbler (1954) lists, in addition to the genera previously mentioned, a species of *Poecilus* and several species of *Bembidion* as destroying seed in nursery beds.

Carabid-caused damage to Douglas-fir seed is similar to that which Kangur (1954) reported as caused by shrews. Seed coat remains of damaged seed have the ragged edges characteristic of shrew damage but the hulls are less broken (fig. 1). The beetle gnaws through the seed coat, generally starting on the flat surface of the seed, and hollows out the endosperm, leaving most of the seed coat intact.

Of several species[2] thus far tested in the laboratory, *Harpalus cautus* Dejean and *Pterostichus vulgaris* L. readily fed on Douglas-fir seed. Hatch (1953) lists *P. vulgaris*, a species introduced from Europe, as occurring mostly around homes and greenhouses, and *H. cautus* as being common in forested areas from British Columbia to Oregon and east to Idaho. He describes *H. cautus*, the species found with the damaged seed in the field, as being 7.3 to 10.4 mm. long, black with piceous tarsi and fuscous antennae (fig. 2). This species, often found under pieces of bark and wood on the ground, when exposed moves rapidly to a new place of shelter.

Limited field tests conducted in June 1957 indicated that carabid beetle damage was not great. Sixteen 20-seed lots were exposed in the field for 10 days. During this period only five seeds, or less than 2% were damaged. Greater loss may occur

[2] Identification of the beetles by courtesy of Dr. Melville H. Hatch.

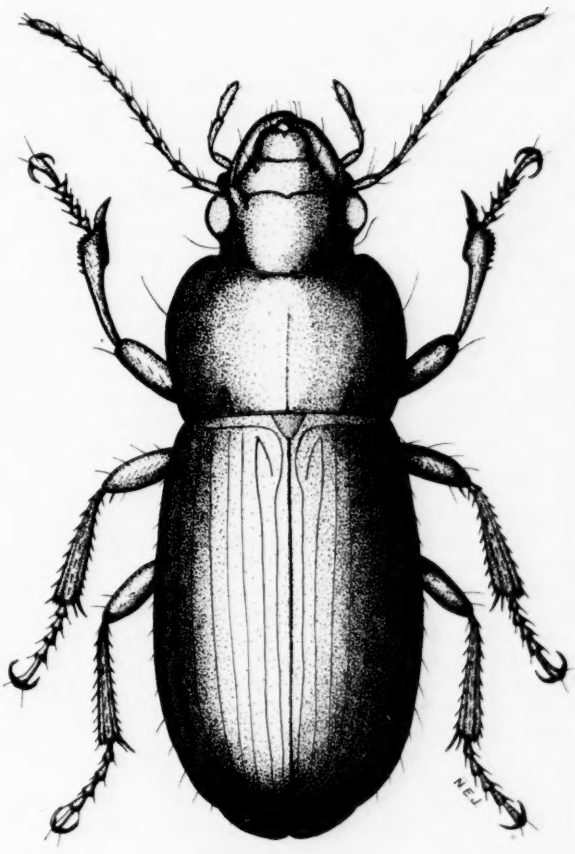


FIG. 2.—*Harpalus cautus* Dejean.

in seed exposed for prolonged periods or at times of increased insect activity.

REFERENCES CITED

Essig, E. O. 1942. College entomology. The MacMillan Co., New York, 900 pp., illus.

Gäbler, H. 1954. Tierische Samenschädlinge der einheimischen forstlichen Holzgewäsche. Neumann Verlag, Radebeul und Berlin. 56 pp. illus.

Hatch, Melville H. 1953. The beetles of the Pacific Northwest. Part I, Vol. 16. Univ. of Washington Press, Seattle. 340 pp. illus.

Imms, A. D. 1948. A general textbook of entomology. E. D. Dutton & Co., New York. 727 pp., illus.

Kangur, R. 1954. Shrews as tree seed eaters in the Douglas-Fir Region. Oregon State Board of Forestry. Res. Note 17. 23 pp., illus.

Nitzsche, H. 1893. Ein neuer Fall von Saatkampbeschädigung durch Laufkafer Forst.—naturw. Zeitschr. 2(48).

Nüsslin, Otto, and L. Rhumbler. 1922. Forstinsektenkunde 3rd. ed. Verlagsbuchhandlung. Paul Parey, Berlin. 568 pp. illus.

Pollination Studies on Low-Bush Blueberries[1]

WILLIAM R. LEE, *University of New Hampshire, Durham*

The low-bush blueberry, *Vaccinium lamarckii* Camp, is native to New Hampshire where its wild berries were once picked by Indians and early settlers. In recent times improved practices in the management of wild blueberries have made them commercially important. Commercial blueberry fields are developed in areas where low-bush blueberries are already established, since a practical method of establishing the plants in new fields has not been developed. Competitive plants are removed by cutting, chemicals, and burning, allowing the spread of blueberry clones already present in the field. Thus a commercial field represents a collection of a large number of genetically different clones. Even though these clones may not be self-fruitful, compatible pollen should always be available due to the presence of a large number of different clones in the field.

Phipps (1930) recorded about 40 different species of insects visiting blueberry blossoms in Maine. The population of native pollinating insects observed in remote areas of New Hamsphire where blueberries are grown probably provides adequate pollination for small fields. However, large fields of several hundred acres produce a large amount of bloom for a short period of time in relation to the number of native pollinating insects. Since the beekeeper can provide large concentrations of honey bees during the blooming period, it is logical to assume that honey bees can economically restore the balance between insect pollinators and blossoms. An experiment was conducted for the purpose of measuring the capability of the honey bee in pollinating the low-bush blueberry.

While there have been numerous pollination studies on high-bush blueberry, *V. corymbosum* L., relatively little work has been done on the low-bush blueberry. Phipps (1930) and Karmo & Kinsman (1954) reported that low-bush blueberry blossoms are dependent upon insects for pollination. For purposes of experimental design it is assumed that the low-bush blueberry is self-unfruitful. This assumption is supported by observations made by E. M. Meader while conducting plant breeding experiments at the University of New Hampshire. It was felt desirable to make comparisons within clones because of the possible large variation among clones.

EXPERIMENTAL DESIGN.—Three pairs of cages 3′×6′×3′ made of 18×14-mesh screen wire were placed over clones of low-bush blueberries. Each pair of cages was placed over two adjacent clones so that a part of each clone was in both cages (see fig. 1). One cage in each pair contained a nucleus (small bee colony). Fruit set was determined by counting the number of blossoms on 10 stalks of each clone in each cage (20 stalks per cage) and then a month later counting the number of developing berries.

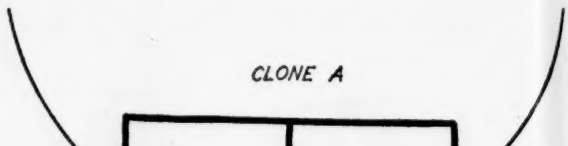


FIG. 1.—Diagram of a pair of cages placed over two adjacent clones of low-bush blueberries. Heavy lines represent cage boundaries while lighter lines mark boundaries of clones.

RESULTS.—The average per cent of fruit set for each clone and cage is shown in table 1. The great difference in fruit set between the portion of the clones with bees and the portion without bees is evidence that insect pollinators are essential and that the honey bee is capable of pollinating low-bush blueberries. Foraging honey bees were frequently observed on blossoms of low-bush blueberries in fields close to colonies. While the environment within cages is not comparable with that outside, if one considers the data in table 1 in connection with the observation that honey bees work low-bush blueberries readily

[1] Published with the approval of the Director of the New Hampshire Agricultural Experiment Station as Scientific Contribution No. 217. Accepted for publication February 7, 1958.

Table 1.—Per cent of low-bush blueberry blossoms setting fruit in cages with and without honey bees.

	CAGE WITH BEES	CAGE WITHOUT BEES
	Cage Pair 1	
Clone A	87.9	0.0
Clone B	55.3	0.0
Average	71.6	0.0
	Cage Pair 2	
Clone C	98.0	11.1
Clone D	86.0	18.0
Average	92.0	14.6[a]
	Cage Pair 3	
Clone E	36.4	0.0
Clone F	43.4	4.7[b]
Average	39.9	2.4
General average	67.8	5.6

[a] One bumble bee gained entrance to this cage for several days.
[b] Several honey bees gained entrance after clone E had stopped blooming.

under field conditions, it is evident that the honey bee is an effective pollinator of this plant.

Unfortunately some bees gained entrance to two cages where attempts had been made to exclude them, thus explaining the partial fruit set on clones C, D, and F. The absence of berries on clones A, B, and E is consistent with the results of Karmo & Kinsman (1954) and Phipps (1930) that the low-bush blueberry is obligate for insect pollination.

One week before harvest the number of berries on each stalk was again counted. In the cages with bees 93.9% of the fruit set matured. In uncaged plots in the same field 95.1% of the fruit set matured. The low-bush blueberry is capable of setting and maturing fruit on a high percentage of its flowers; however, considerable variation exists between clones in the percentage of fruit set.

An analysis of variance showed the variation between clones within cages containing bees to be significant at the 1% level ($F=4.39$, d.f. 3, 54). Since we can assume maximum pollination under these conditions, this variation must be due to physiological differences between clones. A similar variation between clones within a location was noted on uncaged plots at locations scattered throughout a 20-acre field ($F=4.46$, d.f. 4, 32). This large variance between clones shows the advisability of making comparisons within clones when possible and the necessity of sampling a large number of clones when an estimate of fruit set for a field is made.

REFERENCES CITED

Karmo, E. A., and G. B. Kinsman. 1954. The place of honey bees in low-bush blueberry pollination. Nova Scotia Dept. Agric. and Marketing, Mimeographed Circ. Serv. No. 77.

Meader, E. M. 1957. Personal communication.

Phipps, C. R. 1930. Blueberry and huckleberry insects. Maine Agric. Expt. Sta. Bull. **356**: 107–232.

Tests with Dow ET-57 and Bayer 21/199 Against Cattle Grubs in Southwest Louisiana[1]

EDWARD C. BURNS[2] and E. E. GOODWIN[3]

For many years rotenone has been the standard recommendation for controling the cattle grubs *Hypoderma lineatum* (De Vill.) and *Hypoderma bovis* (L.). While this treatment has not been satisfactory, it has been the best available. With the discovery of the effectiveness of the systemic insecticides, Dow ET-57 and Bayer 21/199, a method of control is provided which appears to offer many advantages over rotenone which has to be applied several times in order to obtain effective control. One treatment with the systemics usually gives control, and young larvae are destroyed before they reach the backs of the animals where a large amount of damage normally is inflicted to the tissues and hides.

Table 1.—Effect of time of treatment on grub control. Crowley, Louisiana, 1957.

Date of Treatment	Dow ET-57 (100 Mg./Kg. Bodyweight) No. of Animals	Grubs per Cow[a]	Per Cent Control	Bayer 21/199 (0.5 Gal. 0.75%) No. of Animals	Grubs per Cow[a]	Per Cent Control
June 20	6	2.0	79	6	4.3	56[b]
June 20+August 20	5	0.4	96	6	0.0	100
August 20	6	0.7	93	6	0.0	100
Untreated	4	9.7		4	9.7	100

[a] December 13, 1957.
[b] Heavy rain (1.45 inches) June 20, 1957, beginning 1 hour after treatment.

Since the systemics are capable of destroying the young larvae as they migrate through the tissue, it would seem desirable to treat animals as soon as possible after completion of the oviposition period of the fly. Experiments were designed to investigate timing of treatment of cattle and to determine the relative effectiveness of several rates of Bayer 21/199. In all of the experiments Dow ET-57 was given as a bolus at 100 mg./kg. estimated body weight and Bayer 21/199 was applied as a wettable powder spray at the rate of one-half gallon per animal. Since most of the cattle in the tests belonged to commercial cattlemen and could not be examined as often as desired, data are based on grub counts taken in midDecember at which time largest numbers were thought to be present in the backs of local cattle. Spot checks before and after this date supported this procedure.

In the first of these experiments two groups of animals from the same herd were treated on June 20th with Dow ET-57 and Bayer 21/199, respectively. These groups were divided into halves with one-half receiving a second treatment of the same material and concentration on August 20th. Two more groups received ET-57 or 21/199 on August 20th only. Another group was held as a check. The results of this test are shown in table 1.

Four herds of cattle were used in the second experiment. The dosage of ET-57 was held constant at 100 mg./kg. of bodyweight, but the concentration of 21/199 was varied from 0.25%–0.75%. Results of this experiment are shown in table 2.

In the first experiment effective control was obtained with Dow ET-57 at both dates but control was less effective when the animals were treated on June 20th. This would seem to indicate the need for later treatment. However, the number of animals used was very limited, and results from the second experiment do not support this assumption. Excellent control was obtained with Bayer 21/199 in the early plus late treatment and in the late treatment. Poor results were obtained in the group which received treatment on June 20th only. A heavy rain 1 hour following spraying no doubt was responsible for this.

Results from experiment No. 2 indicate that effective control may be obtained with Bayer 21/199 at 0.25% concentration.

[1] Accepted for publication February 24, 1958.
[2] Department of Entomology, Louisiana State University, Agricultural Experiment Station, Baton Rouge.
[3] Louisiana State University, Rice Experiment Station, Crowley.

Table 2.—Cattle grub experiments. Crowley, Louisiana, 1957.

Herd No.	Date of Treatment	Dow ET-57 (100 Mg./Kg. Bodyweight) No. of Animals	Grubs Per Cow[a]	Per Cent Control	Bayer 21/199 (0.5 Gal./Animal No. of Animals	Per Cent Solution	Grubs Per Cow[a]	Per Cent Control	Control No. of Animals	Grubs Per Cow[a]
1	June 20	16	0.94	91.0	17	0.75%	.20	98.0	31	10.5
2	July 18	18	1.30	83.9	40	.50%	.05	99.4	25	8.1
3	July 26	9	2.20	85.1	23	.50%	.00	100.0	20	14.8
					30	.25%	.13	99.8	20	14.8
4	July 27				35	.50%	.17	96.4	16	5.1

[a] December 10–16, 1957.

House Fly Resistance to Organophosphorus Compounds in Arizona and Georgia[1]

H. F. SCHOOF and JOHN W. KILPATRICK[2]

The initial occurrence of resistance to malathion in the United States by *Musca domestica* L. was reported by Kilpatrick & Schoof (1958) with flies from a dairy at Savannah, Georgia. Other workers (Lindquist 1957, Labrecque & Wilson 1957) have indicated malathion resistance in Florida; while Keiding (1956) and Sacca (In press) respectively showed organophosphorus resistance in Denmark (Diazinon, Bayer 21/199) and in Italy (Diazinon). This note discusses field strains of *M. domestica* from Savannah, Georgia, and Phoenix, Arizona, which are resistant to malathion, Diazinon, and parathion.

SAVANNAH, GEORGIA.—A local chicken ranch (Anderson) reported failure of malathion poison baits and residual treatments in controlling house flies in late August 1957. Observations confirmed this report, with a grill index of 135 flies being obtained on September 10. Application of malathion (2%)–DDVP (0.25%) dry poison bait at a rate of 4 oz./1000 sq. ft. on plywood trays gave no observable reduction in densities, with fly indices on September 15 and 18 being 207 and 131, respectively. On September 18, a residual treatment of a malathion-sugar formulation was made to all potential resting surfaces at a rate of 200:500 mg. of malathion:sugar/sq. ft. The index fell to 69 flies on September 20 but by September 26 rose to 104 flies. At that time, treatment with a 0.1% DDVP-10% sugar liquid bait on cellucotton pads effectively reduced the fly population. Subsequent laboratory exposure of field-collected specimens for 30 minutes to 1-week-old deposits of malathion:sugar (200:500 mg./sq. ft.) and to 8-week-old deposits of DDT (200 mg./sq. ft.) on plywood panels gave 24-hour mortalities of 8% and 2%, respectively. Such field and laboratory data suggested a high level of physiologic resistance to malathion.

The Anderson strain then was colonized without selection pressure and 3-day-old females subjected to topical applications of malathion, Diazinon, and parathion. As the data (table 1) show, the strain had an extremely high resistance to malathion and was less susceptible to parathion and Diazinon than the CSMA (Chemical Specialties and Manufacturers Association) laboratory strain.

Previous history of insecticidal treatment at this ranch revealed the continuous use of a malathion dry bait from 1955 through 1956, a limited installation of parathion impregnated cords in late 1956, and the application of both malathion baits and residual sprays in 1957.

Behavioristic resistance similar to that previously encountered with the Bethesda strain (Kilpatrick & Schoof 1958) also was observed in the Anderson strain.

PHOENIX, ARIZONA.—In August 1957, C. Heard Field (Industrial Fly Control Service) reported that Diazinon applications no longer were giving effective control of house flies in dairy barns in the Phoenix area. House fly pupae were forwarded to Savannah and the strain colonized without selection pressure. Exposure of 3-day-old females for 30 minutes to one-week-old deposits of malathion:sugar (200:500 mg./sq. ft.), Diazinon:sugar (100:250 mg./sq. ft.), and DDT (200 mg./sq. ft.), yielded a mortality of 1% with each toxicant. Topical applications of malathion, Diazinon, and parathion indicated the strain was resistant to all three compounds, and particularly so to malathion (table 1).

The Sharp dairy at which the strain was collected had been treated premises-wide about 66 times (applications of approximately 25 gallons every 2 to 4 weeks) since 1954 with organophosphorus insecticides. In 1954 and 1955, 26 applications of either 2 or 2.5% malathion plus a 12.5% sugar additive provided excellent house fly control. In early 1956 a similar level of control was achieved. However, a decreasing effectiveness of the sprays in early July led to a replacement of the malathion emulsion with a 1% Diazinon-6.25% sugar formulation. The Sharp dairy received seven treatments of Diazinon in 1956, similar periodic applications being continued in 1957 until June when poor control was apparent. Three residual treatments of Diazinon:sugar and one each of Diazinon-DDVP and malathion-sugar from June 19 to July 20 failed to give sustained residual effectiveness. An installation of parathion-Diazinon cords in mid-August reduced fly densities but did not provide satisfactory control.

The level of malathion resistance in the Sharp strain is comparable to that of Bethesda strain after selection by exposure to malathion deposits (Fay *et al.*, in manuscript). Both of these strains appear more susceptible to malathion than the Anderson strain (table 1). Of the three resistant strains, the Sharp strain showed the least susceptibility to parathion and Diazinon.

Table 1.—Twenty-four-hour per cent mortality of three strains of house flies receiving topical applications of malathion, Diazinon, or parathion (25 females per dosage).[a]

Toxicant-Strain	Micrograms of Toxicant per Fly								
	.01	.05	0.1	0.5	1.0	9.0	18	66	72
Malathion									
CSMA[b]	—	0	0	100	—	—	—	—	—
Anderson	—	—	—	—	—	48	68	92	100
Sharp	—	—	—	4	16	60	88	100	
Bethesda-45	—	—	—	—	—	32	92	100	
Diazinon									
CSMA	0	92	100	—	—				
Anderson	0	4	20	100	—				
Sharp	0	0	0	56	96				
Bethesda-45	0	20	60	100	—				
Parathion									
CSMA	56	100	—						
Anderson	0	52	100	100					
Sharp	0	24	52	100					
Bethesda-45	0	80	100	100					

[a] All exposures made during the same 3-day interval. Additional replications with the individual strains at different times gave similar results.

[b] Dosages of 0.2 and 0.3 micrograms/fly gave mortalities of 20 and 36%, respectively.

REFERENCES CITED

Kilpatrick, John W., and H. F. Schoof. 1958. A field strain of malathion-resistant house flies. Jour. Econ. Ent. 51(1): 18–19.

Fay, R. W., John W. Kilpatrick, and G. Clinton Morris, III. Malathion resistance studies on the house fly (In manuscript).

Labrecque, G. C. and H. G. Wilson. 1957. House fly resistance to organophosphorus compounds. Agric. Chemicals 12(9): 46, 47, 147, 149.

Sacca, G. La Resistenza di *M. domestica* Agli Esteri Fosforici in Provincia di Latina. Riv. Parasit. (In Press).

Keiding, J. 1956. Resistance to organic phosphorus insecticides of the house fly. Science 123(2309): 1173–4.

Lindquist, Arthur W., 1957. Effectiveness of organophosphorus insecticides against house flies and mosquitoes. Bull. World Health Organ. 16(1): 33–39.

[1] Accepted for publication February 24, 1958.

[2] From the Communicable Disease Center, Bureau of State Services, Public Health Service, U. S. Department of Health, Education, and Welfare, Savannah, Georgia.

Human Ear Invasions by Adult Scarabaeid Beetles[1]

DARRELL R. MADDOCK and CLYDE F. FEHN[2]

The unusual occurrence of 186 cases of adult June beetles (Scarabaeidae) entering the ears of sleeping boys was observed

[1] Accepted for publication March 24, 1958.

[2] From the Communicable Disease Center, Bureau of State Services, Public Health Service, U. S. Department of Health, Education, and Welfare, Savannah, Georgia.

at the Boy Scout Jamboree at Valley Forge State Park, Pennsylvania, in July 1957. Published reports of a similar nature are rare. Hallock (1936) mentions adult beetles invading ears and impairing hearing through ear drum injury. Metcalf *et al.* (1951) state that adult annual white grubs (*Ochrosidia villosa* Burmeister) "have occasionally caused personal injury by burrowing in the external ear of sleeping persons." Such invasions are relatively common in the Middle West where June beetles are extremely abundant, according to R. L. Metcalf (personal communication), but reports have not been published.

The 3000-acre State Park is comprised of gently sloping low hills primarily covered with sod. The majority of the 52,580 Jamboree participants slept on the ground in two-man tents, the remainder using cots.

During the first two nights of the encampment, all ear invasions were by the northern masked chafer (*Cyclocephala borealis* Arrow).[3] The Asiatic garden beetle (*Autoserica castanea* Arrow)[3] began to appear on the third night and subsequently was the only species involved. *C. borealis* is about 10 mm. long and 6 mm. wide while *A. castanea* is slightly smaller. Information obtained from the U. S. Department of Agriculture[4] indicates that *A. castanea* and *C. borealis* have been distributed widely in the northeastern United States since at least 1936. These data give no indication of regularity in yearly cycles of abundance. Hallock (1936) observed that low populations are associated with preceding summers of deficient rainfall. Rainfall in the summer of 1956 was average but 1949 was very dry. This perhaps explains why no problem developed during the 1950 Boy Scout Jamboree, also held at Valley Forge during July.

The presence of a beetle in the ear canal was extremely painful because the tibial spines pierced the delicate skin of the canal as the beetle forced its way in—generally so deeply it could not be seen without an otoscope. Removal was likewise accompanied by considerable pain and slight bleeding. The effects apparently were transitory with very few cases of secondary infection.

The number of cases reported (186) is an adjusted composite of those treated by the U. S. Army Field Hospital[5] (142 cases) and the Health Lodge medical staffs. After the first few days all victims were referred to the hospital, arriving between 11:00 P.M. and 1:30 A.M. A smaller number occurred near sunrise. The groupings were related to the late evening emergence of the beetles from the turf and their return at daybreak. With two exceptions, in which beetles flew directly into the ears of two boys standing in lighted areas, all boys were sleeping on the ground when the invasions occurred. No cases were reported in persons sleeping on cots.

Counts were made of the number of beetles between 11:00 P.M. and midnight in six illuminated commissary tents initially selected at random. The side curtains of these 3000 sq. ft. tents were kept partly open for ventilation. The first two nights the beetles "at light" were largely *C. borealis*. Subsequently, *A. castanea* increased in number and *C. borealis* practically disappeared, a trend in agreement with the reversal in the occurrence of each species in ears.

The cases were widely distributed, occurring in various parts of the camp on different nights. From the data, it appears that populations of *C. borealis* were declining at the beginning of the Jamboree and that *A. castanea* reached a peak of activity on the night of July 13 (fig. 1). The number of cases was related to the general beetle abundance "at light". Night temperatures (recorded at the U. S. Weather Bureau at Philadelphia International Airport, 19 miles away) ranged from 59 to 80° F. Fewer cases occurred on the coolest nights (10th and 15th) but the relationship with temperature at other times was inconsistent.

Emergency control measures were considered after the occurrence of 51 cases on the night of July 13. Insecticidal treatment of over 25,000 two-man tents and 1200 acres of turf in the camping area was impractical. Therefore, it was recommended that mechanical means be utilized to exclude the beetles from the ears. Ear plugs of cotton or toilet tissue or Boy Scout neckerchiefs

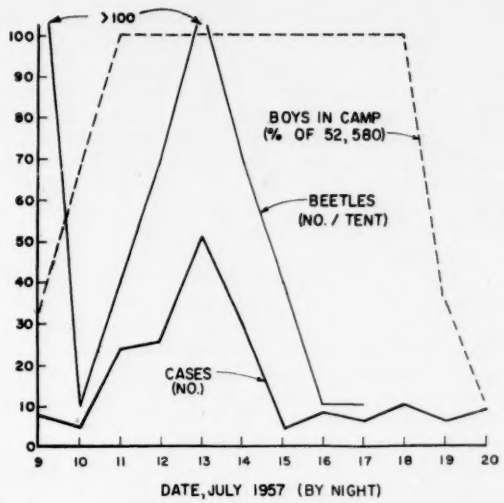


FIG. 1.—The number of ear invasions by adult beetles compared with beetle abundance at light.

wrapped around the head to cover the ears were used by a small portion of the camp population. In one instance, a beetle was removed from an ear previously plugged with cotton. Data were insufficient to evaluate the effectiveness of the preventive measures.

The incidence of cases was sufficiently great that preventive measures warrant consideration in planning similar encampments.

REFERENCES CITED

Hallock, H. C. 1936. Life history and control of the Asiatic garden beetle. U. S. Dept. Agric. Circ. **246.**

Metcalf, C. L., W. P. Flint, and R. L. Metcalf. 1951. Destructive and useful insects. 3rd Ed. New York, N. Y.: McGraw-Hill, 1071 pp.

[3] Identifications were made by Mr. O. L. Cartwright, Associate Curator, Division of Insects, U. S. National Museum, Washington, D. C.

[4] The authors wish to thank Dr. P. W. Oman and Mr. J. W. Gentry of the Agricultural Research Service, U. S. Department of Agriculture, for information concerning the biology of the beetles.

[5] The authors wish to thank Major Harry N. Jones, M.D., U. S. Army, for his assistance in this study. Major Jones and his associates are preparing an article on treatment techniques for publication in a medical journal.

Technique for Testing Insecticide Deposits with Newly Hatched Codling Moth Larvae[1]

M. M. BARNES, *University of California Citrus Experiment Station, Riverside*

The development of suitable laboratory testing procedures is a process of selecting techniques which eliminate as many variables as possible and provide results which are reproducible within desired limits. The procedures described were designed to test the effectiveness of initial or weathered deposits of insecticides against newly hatched larvae of the codling moth, *Carpocapsa pomonella* (L.), using a reasonably small number of larvae and providing results which are reproducible within reasonably narrow limits. Many features of this procedure are based on a technique developed by Carman & Fleschner (1944) for insecticide studies with the oriental fruit moth, *Grapholitha molesta* (Busck), and used in separate studies with the codling moth.

Apple fruits used in laboratory tests are washed in a solution of trisodium phosphate, water-rinsed, and air-dried. As the fruit rotates (35 r.p.m.) on a turntable, spray is applied during 3

[1] Paper No. 1022, University of California Citrus Experiment Station, Riverside, California. Accepted for publication April 7, 1958.

rotations with a DeVilbiss air-pressure sprayer. The spray application as applied results in very little run-off. When the spray deposits are dry, six cells are affixed to the equator of each fruit (fig. 1) by applying a molten mixture of paraffin and beeswax (50-50) with a medicine dropper around the inside of the cell where it contacts the fruit surface. The insecticide deposit which is assessed is therefore that on the cheek of the fruit. This deposit is more uniform than that on the other surfaces of the fruit, and there is less variation from test to test. The cells currently in use are $\frac{3}{4}$-inch sections of 1-inch aluminum tubing. (Glass tubing also may be used.) Prior to use, the metal cells are dipped, 200 or 300 at a time, in the paraffin-beeswax mixture; after use, they are cleaned by melting and draining off the excess wax and then dipping the cells in carbon tetrachloride. Slightly higher "natural mortality of larvae results in using the metal cells unless they are dipped in wax. Glass cells do not have this disadvantage, but unless cut very evenly they are difficult to affix to the apple without leaving an escape hole.

Codling moth populations are reared and eggs are obtained on waxed paper, as described by Dickson *et al.* (1952). Tests may be run at any time of the year, since rearing is continuous. Egg sheets are held at 26° C. until they reach the blackhead stage. Small triangles of waxed paper, each bearing one blackhead-stage egg may be rapidly cut from the egg sheet with a razor blade, and these units may be used for infestation. If eggs are used which have not reached the blackhead stage, a lower percentage of hatch is usually encountered and a smaller total number of larvae enter the experiment than anticipated. Newly hatched larvae may also be used for infestation (Steiner 1939). In infesting the fruit of any one run, it is relatively important that this be done in replicate fashion, i.e., one fruit of each treatment or untreated control at a time, in series. This tends to eliminate the bias in selection of eggs or larvae for infesting which exists if all of the fruit of one treatment are infested first, then the untreated control fruit, etc. The tendency in infesting fruit is to select first the larger eggs that are in the late blackhead stage, or the more active larvae; the last fruit infested thus receives smaller eggs or less active larvae. If not corrected, this tends toward bias, in that infestation of the last lots of fruit may be with larvae of lower vigor and eggs of lower viability. Various susceptibilities and various abilities to enter fruit are selected evenly if fruits are infested as replicates.

After the cells are attached to the fruit, a triangle of waxed paper bearing an egg, or a single newly hatched larva, is placed on the inner wall of each cell. The cell is closed by a square of broadcloth held in place by a cork ring (see fig. 1). If fumigation effects are a factor, these may be mitigated by omitting the closure and ringing the top of the cell with a barrier of a suitable sticky material. The newly hatched larva crawls to the circle of the fruit surface thus enclosed and attempts to enter. If the larva dies, it is readily recovered as it cannot fall off the fruit. Isolation of the larvae provides another advantage over free infestation since it prevents entrance into bites or "stings" resulting from activities of other larvae.

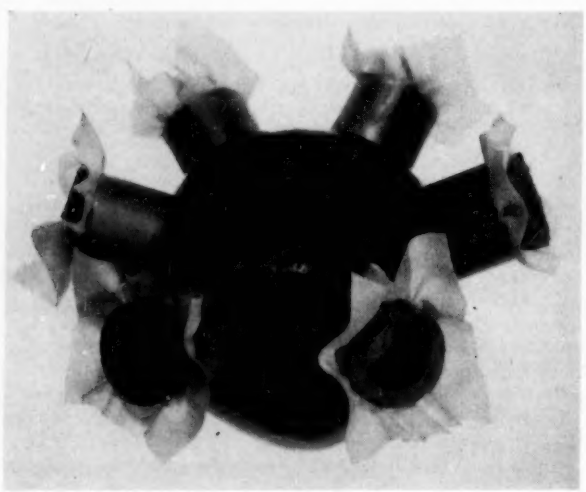


FIG. 1.—Fruit with metal cells attached to isolate codling moth larvae and to provide assay of deposit on the cheek of the fruit during each run.

Temperature is a factor in the ability of codling moth larvae to enter unsprayed fruit (Hough 1934). This variable is controlled by placing newly infested fruit in a constant-temperature cabinet held at 26° C. Relative humidity in this cabinet is maintained at 85% to 90%, since it has been observed that high humidity favors good hatch of eggs on waxed paper.

Selection of the number of eggs or larvae to be used for any one treatment and control run is a matter of counterbalancing the need for reliable results against the factors of time and effort. Mortality of eggs is usually under 10%. An entry is defined as penetration of the larva into the fruit for a distance of over one quarter inch.

Table 1 provides information on the day-to-day reproducibility of this procedure, using Rome Beauty apples and about 40 larvae per test. In these tests a dosage of a standard DDT wettable powder was used, which is the approximate LC 50 of the standard laboratory strain of codling moth. This laboratory strain is not resistant to DDT and, reared continuously, has maintained the same LC 50 since 1948. It should be noted that the data in table 1 are from 10 tests on different days, using different egg sheets in each test, and, as previously indicated, using about 40 larvae for each test. Under these conditions the average mortality from the standard dosage of the standard DDT wettable powder was 45% with a standard deviation of 9%. Greater precision can readily be achieved if desired by using a larger number of larvae. In individual laboratory comparisons pertaining to resistance to DDT of field-collected strains as compared with the standard strain for, example, at least 100 eggs were used on 17 fruits. Dosage-mortality relationships obtained with 100 eggs for each point are quite symmetrical.

Untreated fruits infested with each strain are included in each run. This provides a control for ability of the larvae to enter untreated fruit. Occasionally, the percentage of entries on the untreated fruit with the standard strain will be unaccountably low. Results of runs when the proportion of entries on untreated fruit with the standard strain falls below 80% are discarded.

Table 1.—Reproducibility of results when the cell technique is used with relatively few newly hatched codling moth larvae on different days.

TEST NO.[a]	NUMBER OF LARVAE	PER CENT REDUCTION OF FRUIT ENTRIES[b]
1	39	46
2	40	47
3	38	39
4	36	56
5	40	55
6	41	30
7	35	48
8	40	35
9	39	43
10	42	54
Mean		45
Standard deviation		8.7
Standard error		2.6

[a] In each test seven Rome Beauty fruits were sprayed with 1.2 gm. per liter of a standard 50% DDT wettable powder. Each fruit was infested with six blackhead-stage eggs isolated in attached metal cells.

[b] As calculated by Abbott's formula, using percentage of entries on unsprayed fruit as a control.

This procedure offers the advantages of: (1) confining individual larvae and thus avoiding fruit entry through "stings" caused by other larvae; (2) preventing loss or escape of larvae; (3) assaying only the deposit on the equator of the fruit, which is less variable than other areas; and (4) by control of these variables, providing reproducibility within reasonably narrow limits with the use of a reasonably small number of larvae.

REFERENCES CITED

Carman, G. E., and C. A. Fleschner. 1944. Laboratory tests on the oriental fruit moth with special reference to DDT. Jour. Econ. Ent. **37**(1): 122–3.

Dickson, R. C., M. M. Barnes, and C. L. Turzan. 1952. Continuous rearing of the codling moth. Jour. Econ. Ent. **45**(1): 66–68.

Hough, W. S. 1934. Colorado and Virginia strains of codling moth in relation to their ability to enter sprayed and unsprayed apples. Jour. Agric. Res. **48**: 533–53.

Steiner, L. F. 1939. The laboratory-field method for testing codling moth insecticides. U. S. Bur. Ent. and Plant Quar. **E-488.**

Triatoma sanguisuga Infesting a Bedroom in Decatur, Georgia[1]

HAROLD G. SCOTT, *Communicable Disease Center, Atlanta, Georgia*

An infestation of bloodsucking conenose bugs, *Triatoma sanguisuga* Leconte, (Hemiptera: Reduviidae) (fig. 1) in an upper income home in Decatur, Georgia, was investigated in July 1956. The bugs were found to be harboring in and around the bed, particularly between the mattress and the mattress cover. All furniture, including the bed, was made of wood and presented an unusually large number of wood-to-wood joints. This factor may have encouraged development of the infestation. The bedroom was occupied by a married couple. The man had at least three bites attributable to the bugs. The woman apparently had not been bitten. No other rooms in the house were infested. The infestation was eliminated by using a 5% DDT suspension applied to possible harborages. Verification of the determination was made by Dr. Harry D. Pratt.

FIG. 1—*Triatoma sanguisuga* Leconte, dorsal view.

[1] Accepted for publication April 4, 1958.

Control of Iris Borer with Phosphate Insecticides[1]

JOHN C. SCHREAD, *Connecticut Agricultural Experiment Station, New Haven*

Control of iris borers (*Macronoctua onusta* Grote) in the folds of iris leaves and in the lower parts of the fans and flower stems has until recently been difficult. Felt (1912) first reported the probability of controlling newly hatched larvae with arsenate of lead. Dietz (1928) obtained 90 to 95% control of borers with arsenate of lead plus fish oil soap. Ries (1929) achieved similar results with the same combination of materials in addition to arsenate of lead plus casein. The use of pyrethrum (Dietz 1935) and corrosive sublimate (McKinney 1924) as dips did not prove useful under field conditions. Cory (1928), however, reported 92% kill of iris borer pupae with carbon bisulphide emulsion soil treatments. Rotenone (Dietz 1935) was reported to have given more complete control of newly hatched larvae in the spring than arsenate of lead. The use of DDT for this purpose was first attempted in early 1946 by Lowry & Fraim (1947). They included sulphur and fermate with the insecticide and applied the formulation in both dust and spray form with a sticker and spreader added to the latter. The results of their experiments indicated a marked decrease in borer population. Schread (1957) demonstrated the effectiveness of Thimet and Trithion in destroying the borers in the rhizomes and soil. Demeton gave somewhat less control.

MATERIALS AND METHODS.—Experiments on control of iris borer in 1957 were carried on during May soon after the larvae had hatched and again during July when they were about one-half grown. Phosdrin 95%, Bayer 19639 26.5%, and Dibrom 42%[2] emulsions were used in the tests undertaken on May 22 when the borers varied in length from $\frac{3}{16}$″ to $\frac{5}{8}$″. The materials were sprayed onto both sides of the iris plants at dilutions of 1 to 400 and 1 to 800 of the formulation. Triton B-1956 was added to the sprays at a dilution of 1 to 800. All treated plants and checks were 2-year-old transplants growing in 3-foot beds averaging 18 feet in length. Twelve varieties of iris varying in height from 6″ to 24″ were growing in the beds. An examination of 50 plants indicated 63.6% borer infestation at the time of treatment. A 3-gallon hand pressure sprayer was used to make the treatments, applying an average of 1 quart of complete spray to 30 sq. feet of iris bed.

Phosdrin and Bayer 19639 in addition to Sytam[3] 42% emulsion were used on additional borer-infested plants on July 8, at dilutions of 1 to 800 and 1 to 1600 of the formulation with Triton B-1956 added at 1 to 1600. Because of its non-systemic action Dibrom was not included in these tests, instead Sytam was substituted. To avoid possible foliage injury at the time of treatment when extremely hot, dry weather prevailed, lower dilutions of the materials were, in part, employed in July than in May. The plants were growing in 12-foot beds. A pretreatment exam-

[1] Accepted for publication April 7, 1958.

[2] O,O-Dimethyl-O-(1,2-dibromo-2,2-dichloroethyl) phosphate.

[3] Trade name for an emulsion concentrate containing 4 pounds of schradan per gallon, sold by Pennsait Chemicals Corporation.

ination of 12 plants taken at random from the beds indicated a total of 14 borers, three of which were in one plant. All were at or near the base of the plants and varied from ¾″ to 1″ in length. None were found in the rhizomes. The method of spraying and the quantity of spray used per treatment was the same as in the May experiments.

Sampling the treated and untreated check plots for data on control was done by cutting all of the infested iris fans close to the rhizomes and separating the leaves for examination. All leaf folds were opened and the leaf stems and flower stalks were split through the centers with a knife to determine the status of the larvae boring therein. In addition all rhizomes in the July treatments were raised out of the soil and dissected. This was supplemented by probing and hand sifting the soil where they were growing. Inasmuch as rhizomes do not become infested until early summer, they were not examined in the beds sprayed in May.

RESULTS AND DISCUSSION.—Table 1 gives the results of the May and June treatments to control iris borer. It is clear that

Table 1.—Control of iris borer with systemic insecticides sprayed on iris foliage. 1957.

INSECTICIDE AND DILUTION OF FORMULATION	NO. PLANTS EXAMINED	BORERS		PER CENT KILLED
		Dead	Alive	
Treatments made May 22—Data taken May 27				
Phosdrin 1–400	60	49	0	100
Phosdrin 1–800	65	40	4	90.9
Bayer 19639 1–400	60	20	7	74.4
Bayer 19639 1–800	60	14	22	38.8
Dibrom 1–400	60	40	5	88.8
Dibrom 1–800	60	18	12	60.0
Untreated check	50	1	32	
Treatments made July 8—Data taken July 12				
Phosdrin 1–800	8	6	0	100
Phosdrin 1–1600	9	6	0	100
Bayer 19639 –800	13	1	2	33.3
Bayer 19639 1–1600	15	1	8	11.1
Sytam 1–800	20	5	4	55.5
Untreated check	25	0	18	

Phosdrin was the best material used for the control of young iris borers in May when they were feeding in shallow mines or exposed on the upper parts of the iris foliage. Moreover, Phosdrin was outstanding in its performance in controlling the half-grown borers in July when they were feeding within the lower parts of the iris fans and flower stems. Dilutions of Phosdrin within the range used in the experiment appeared to make no substantial difference in its efficiency. Because of its high rate of efficiency as a contact insecticide, Dibrom gave good control of young borers in May. Bayer 19639 was reasonably effective at the strongest dilution used in May, but gave poor results at the lowest dilution. Furthermore, because of less efficient translocation of Bayer 19639, when used as a foliage spray, control of iris borers in July when they were confined within the lower parts of the iris plants was poor. Sytam, which is a slow acting systemic, gave poor control of borers in July.

REFERENCES CITED

Cory, Earnest N. 1928. Insect investigations. Maryland Agric. Expt. Sta. Bull. **298**: 186.

Dietz, Harry F. 1928. The status of our knowledge of the iris borer. Amer. Iris Society Bull. **26**: 20–24.

Dietz, Harry F. 1935. The iris borer. Amer. Iris Society Bull. **55**: 74–78.

Felt, E. P. 1912. Iris borer. 27th Report of New York State Entomologist 52–54.

Lowry, F. P., and I. W. Fraim. 1947 April. The use of DDT in control of iris borer. Amer. Iris Society Bull. 105: 64–66.

McKinney, E. P. 1924. The iris borer. Amer. Iris Society Bull. 11: 31.

Ries, Donald T. 1929. Two iris insects. Amer. Iris Society Bull. **32**: 27–46.

Schread, J. C. 1957. Iris borer and its control. Connecticut Agric. Expt. Sta. Circ. **202**: 1–7.

Insects Attracted to Light Traps Placed at Different Heights[1]

S. W. FROST, *The Pennsylvania State University, University Park*

Previous studies on insect catches from light traps placed at different levels have been extremely limited. Herms (1947) operated three traps in a vineyard at 7-, 12- and 15-foot levels for 2 nights. He noted little difference in the catches from these traps, but observed that the Homoptera in general, and the Cicadellidae in particular, preferred the higher traps. Ficht & Hienton (1941) operated 12 traps in a corn field at 5-, 10- and 15-foot levels for a single night. The lower traps caught the largest number of European corn borer moths.

Tests were conducted at or near State College, Pa., using the Pennsylvania insect light trap (Frost 1957), equipped with 15-watt black light fluorescent lamps. These traps were placed one above the other and the heights at which they were hung are designated by the distance from the ground to the bottoms of the traps.

The data have been considerably condensed to reduce the size of table 1. The midges include species of the families Chironomidae. Cecidomyidae, Mycetophylidae and Psychodidae. All other groups were separated to families although not all are included in table 1. Those designated as "other Nemocera" include Anisopidae and Culicidae. The Brachycera include Drosophilidae, Lonchopteridae, Boboridae, Empididae and Anthomyidae. "Other Coleoptera" include Curculionidae, Elateridae, Bostrichidae and Scarabaeidae, exclusive of Phyllophaga. "Other Lepidoptera" include Pyralidae, Crambidae, Geometridae, Arctiidae and Sphingidae. None of these were taken in sufficient numbers to warrant including them separately.

Tests 1 and 2 were conducted at State College where low growing foliage partially shielded the traps hung at the two lower levels. Test number 3 was conducted in an open area unobstructed by foliage.

The large numbers of Coleoptera taken in test number 3 were mostly Hydrophilidae, *Philhydrus ochraceus* Melsh. These minute beetles were sifted from the bulk of the larger insects and counts were made of a weighed sample from which totals were calculated.

RESULTS.—The tests, conducted for 29 nights, yielded sufficient data to draw some definite conclusions. The majority of the insects were taken in traps at the lower levels. This is particularly true of the midges, Tipulidae, Cicadellidae, Miridae, Phyllophaga, Microlepidoptera, Micro- and Macrotrichoptera. In test 3, 3635 *Aphodius*, not shown in table 1, were taken. Of these 2389 came to the lower trap.

The Brachycera, Coleoptera in general, and the Macrolepidoptera showed little difference in selecting traps at different levels.

The smaller insects were usually attracted to traps placed at lower levels and the larger insects were attracted to all levels.

It is difficult to explain why the trap in the middle position

[1] Authorized for publication on March 31, 1958, as paper No. 2244 in the Journal series of the Pennsylvania Experiment Station. Accepted for publication April 7, 1958.

Table 1.—Insects from light traps at different heights.

	TEST NO. 1 13 NIGHTS MAY 7–31			TEST NO. 2 8 NIGHTS MAY 30–JUNE 6			TEST NO. 3 8 NIGHTS JUNE 19–27		
	Height 4′11″	Height 7′9″	Height 10′9″	Height 2′8″	Height 6′10″	Height 11′	Height 6′3″	Height 10′5″	Height 14′5″
Tipulidae	127	96	90	103	107	79	572	379	459
Midges	3,025	2,085	1,690	10,131	5,445	5,202	9,212	4,301	5,871
Other Nemocera	42	28	25	34	46	16	298	264	274
Brachycera	1,412	1,258	1,272	848	570	591	14,931	5,013	22,770
Aphididae	406	343	411	637	953	1,315	31	95	46
Cicadellidae	1,520	1,043	710	2,275	2,234	1,377	18,573	5,924	22,205
Miridae	190	71	45	224	113	86	22,131	7,875	13,484
Hemip.-Homoptera	11	17	15	21	34	38	712	306	1,638
Carabidae	205	241	219	124	134	134	5,919	3,832	7,913
Staphylinidae	197	270	234	57	68	58	2,872	958	1,980
Phyllophaga	13	3	3	21	4	4	305	94	27
Other Coleoptera	279	361	311	287	404	445	290,477	53,448	86,092
Tortricidae	94	62	12	176	135	101	261	238	197
Noctuidae	942	751	689	572	545	502	1,042	940	949
Microlepidoptera	306	109	138	449	385	304	3,652	1,845	1,986
Other lepidoptera	288	199	129	248	157	129	1,093	948	618
Macrotrichoptera	158	68	53	741	623	468	9,232	3,283	2,227
Microtrichoptera	43	11	24	111	67	48	1,895	458	757
Ephemerida	16	11	7	120	29	14	84	4	2
Hymenoptera	96	70	43	57	59	56	9,834	1,453	998
Other insects	591	465	108	120	8	31	76	97	175
Totals	9,961	7,562	6,228	17,356	12,120	10,998	393,202	91,755	170,688

sometimes attracted fewer insects than either the upper or lower trap. The presence of species not identified may account for this.

It is evident that aphids were captured more abundantly in tests 1 and 2 where nearby foliage was present. On the other hand, the Phyllophaga responded more freely to traps unobstructed by foliage.

REFERENCES CITED

Herms, W. B. 1947. Some problems in the use of artificial light in crop protection. Hilgardia **17**(10): 364, 366.

Ficht, G. A., and T. E. Hienton. 1941. Some of the more important factors governing the flight of European corn borer moths to electric traps. Jour. Econ. Ent. **34**(5): 603.

Frost, S. W. 1957. The Pennsylvania insect light trap. Jour. Econ. Ent. **50**: 287–92.

Rapid Determination of Sex in *Rhagoletis completa* Cresson[1]

H. R. MOFFITT, *University of California Citrus Experiment Station, Riverside*

Sex determination in *Rhagoletis completa* Cresson, the walnut husk fly, has previously been based on the presence of the ovipositor, the more pointed abdomen, and the slightly larger size of the female (Boyce 1934). In the original description (Cresson 1929), no differences between male and female were noted.

The size difference between the sexes is variable, and differentiation of sex on this basis alone is difficult. Frequently, the ovipositor and tip of the abdomen are obscured because of the quick movements of live flies and the drying and curling under of the abdomen of dead flies.

During the 1957 season, numerous trials involving the determination and counting of the sexes of captured walnut husk flies were conducted. While these data were being recorded, it was observed that the male is much darker in appearance than the female. To verify this observation, samples of flies representing field populations in Los Angeles, Riverside, and San Bernardino counties were brought into the laboratory.

The sex of the flies was first determined on the basis of the presence of the ovipositor in the female. These same flies were then separated on the basis of color differences between males and females. The two bases of differentiation of sex agreed very well, as only one female, whose sex had been first determined by color, required examination under the microscope to verify its sex. The total number of flies examined was 810; of these, 420 were females and 390 were males.

In color, the male differs from the female primarily in having the coxae and femora black. The coxae and femora of the female are tawny to light brown; never black. The pleura of the male may be entirely black or predominantly black, with a few light- to dark-brown areas on the lower portions. Female walnut husk flies may be entirely light- to dark-brown or predominantly brown, with the upper portion black and a few black areas on the lower portions.

From the lateral aspect, the general appearance of the male is black, while that of the female is light to dark brown. From the dorsal aspect, the two sexes can be separated on the basis of the black femora of the male. The black coloration is not as well developed in teneral specimens as in more mature walnut husk flies, but enough black is evident on the coxae and femora to permit separation on this basis.

The use of this color difference between the male and female of *Rhagoletis completa* greatly facilitates sex determination, both in the field and in the laboratory, particularly when the ovipositor has become obscured in some manner. In differentiating between the sexes on the basis of color, a quick glance suffices and replaces the need for examination of many flies beneath the microscope.

REFERENCES CITED

Boyce, A. M. 1934. Bionomics of the walnut husk fly, *Rhagoletis completa*. Hilgardia **8**(11): 363–579.

Cresson, Ezra T., Jr. 1929. A revision of the North American species of fruit flies of the genus *Rhagoletis* (Diptera: Trypetidae). Trans. Amer. Ent. Soc. **55**: 401–14.

[1] Accepted for publication April 8, 1958.

Control of Citrus Whitefly on Gardenia[1]

H. H. TIPPINS, *Georgia Experiment Station, Experiment*

The citrus whitefly, *Dialeurodes citri* (Ashm.), is the most destructive insect pest of Cape Jasmine (*Gardenia*) in Georgia. Infestations are usually light during the spring but increase to serious proportions in the late summer. Most gardenia plants are more or less covered with sooty mold, a fungus growth associated with whitefly excretions, by September. Plants thus affected present an unsightly appearance and are in a weakened condition. Fewer and poor quality flowers are produced on plants weakened by whitefly attack. There is also some evidence (English & Turnipseed 1940) that plants infested with whiteflies are much more susceptible to cold injury than healthy plants.

The citrus whitefly also attacks chinaberry, crapemyrtle, ivy, privet, osage-orange, persimmon, prickly ash, and many other plants. Chinaberry and privet seem to be the major source of whitefly buildup in Georgia. It has been estimated that a good-sized chinaberry tree will produce an average of between 25 and 50 million whiteflies each season (Wilson & Berger 1937).

Current recommendations for the control of whiteflies include oil emulsions, oil emulsions plus rotenone, parathion, and malathion (Yerkes *et al.* 1940, Quayle 1938, English & Turnipseed 1940, Weigel & Baumhofer 1948, Jordan 1954). The use of oil emulsions is limited to periods of relatively mild weather in that extreme temperatures, either high or low, closely following oil sprays may result in damage to the plants. Parathion and malathion, on the other hand, are not highly effective at temperatures lower than 50 degrees F.

The purpose of the work reported herein was to determine whether whiteflies could be controlled with systemic insecticides and to determine the effect of several other insecticides on the control of the whiteflies at low temperatures. Cold injury to the plants following insecticide treatment was also recorded.

PROCEDURE.—The plants used in these tests were *Gardenia jasminoides* var. *radicans* in individual 6-inch clay pots. The plants had an average spread of about 12 inches and were 6 inches in height. Two such potted plants were used in each treatment. The treatments were replicated four times in Test I and six times in Test II. The treatments were evaluated for whitefly control in each test by collecting two leaves from opposite sides of each plant. The percentage mortality of the whiteflies was determined by examination under a microscope. The plants were held in a lathhouse and were irrigated as necessary. Precautions were taken to avoid wetting the foliage during irrigation.

The treatments in Test I were as follows: 2% granulated Thimet at ½ teaspoonful per pot, 2.5% granulated Bayer 19639 at ½ teaspoonful per pot, 10 cc. of demeton emulsifiable concentrate per gallon of water at ½ pint per pot. Thiodan emulsifiable concentrate at 10 cc. per gallon of water and plants sprayed to the runoff stage and an untreated check. The granulated materials were applied to the surface of the soil in the pots and watered in with one half pint of water. Mortality counts were made 40 days after treatment. All treatments were applied on September 26, 1957.

The treatments in Test II included sprays of the following materials and rates: oil emulsion 4½ tablespoonfuls per gallon, Thiodan 20 cc. per gallon, demeton 1 teaspoonful per gallon, malathion 1 teaspoonful per gallon, and an untreated check. The plants were sprayed to the runoff stage with a compressed-air sprayer. The treatments were applied on November 7, 1957. Whitefly mortality was determined and cold injury was rated 40 days after treatment. Cold injury was rated on a 0 to 4 scale with 0 representing no damage and 4 complete defoliation.

RESULTS.—Meterological data for the period of Test I show that there was a total of 3.47 inches of rainfall and that the minimum temperature (recorded on November 28 and 29) was 31 degrees F.

The results of the test are presented in table 1. The demeton gave significantly better control than was obtained by natural means on the check plots. Mortality from natural causes on check plants averaged 27.9%. Mortality in treatments receiving demeton, Thimet, or Bayer 19639 did not differ significantly.

Table 1.—The effectiveness of several insecticides against whiteflies on Cape Jasmine. Experiment, Georgia, 1957.

TREATMENT AND PERCENTAGE MORTALITY[a]				
Check	Thiodan	Bayer 19639	Thimet	Demeton
27.9	32.2	48.6	61.8	80.2

[a] Means not underscored by the same line differ significantly at P = 0.05.

Table 2.—Control of whitefly on Cape Jasmine. Experiment, Georgia, 1957.

TREATMENTS AND PERCENTAGE MORTALITY[a]				
Check	Thiodan	Demeton	Malathion	Oil Emulsion
14.7	33.2	34.6	59.3	61.7

[a] Means not underscored by the same line are significantly different at P = 0.01.

Table 3.—Cold damage to Cape Jasmine following insecticide treatments for the control of whitefly. Experiment, Georgia, 1957.

TREATMENTS AND COLD DAMAGE RATING[a]				
Malathion	Demeton	Thiodan	Oil Emulsion	Check
0.75	1.00	1.75	2.00	2.17

[a] Means not underscored by the same line are significantly different at P = 0.05.

During the period of Test II, there was a total of 9.96 inches of rainfall and the minimum temperature, 12 degrees F., occurred on December 12. Sub-freezing temperatures were recorded on 12 of the 40 days.

The results of Test II (table 2) show that all of the insecticidal treatments gave highly significant control of the whiteflies. There was less natural mortality in this test, the mean mortality in the check plots being 14.7%. Oil emulsion and malathion gave about the same degree of control and were significantly better than all other treatments.

Table 3 shows that control of the whitefly resulted in an increased plant resistance to cold damage. With the exception of oil emulsion, all of the insecticidal treatments tended to reduce cold damage in the same order that they controlled the insects. Oil emulsions are known to accentuate cold injury.

These data show that malathion is about equal to oil emulsion, and that some of the systemic insecticides may be useful in whitefly control. Control of heavy infestations of the whitefly results in plants that are more cold resistant.

REFERENCES CITED

English, L. L., and G. F. Turnipseed. 1940. Control of the major pests of the satsuma orange in South Alabama. Alabama Expt. Sta. Bull. **248**: 17–21.

[1] Paper No. 333; Journal Series, Georgia Experiment Station, Experiment. Accepted for publication April 7, 1958.

Jordan, C. R. 1954. Control of some common pests of ornamental plants. Georgia Agric. Ext. Serv. Ent. Leaflet No. 8.

Quayle, Henry J. 1938. Insects of citrus and other subtropical fruits. pp. 181–6. Comstock Publishing Co., Inc., Ithaca.

Wiegel, C. A., and L. G. Baumhofer. 1948. Handbook on insect enemies of flowers and shrubs. U. S. Dept. Agric. Misc. Publ. **626,** p. 43.

Wilson, J. R., and E. W. Berger. 1937. Citrus insects and their control. Florida Agric. Ext. Serv. Bull. **88,** pp. 47–50.

Yerkes, G. E., F. L. Mulford, Lucia McCullough, and F. F. Smith. 1940. Gardenia culture. U. S. Dept. Agric. Leaflet **199,** pp. 6–7.

A Hymenopterous Parasite New to Oklahoma[1]

E. A. WOOD, JR., *Entomology Research Division, Agr. Res. Serv., U.S.D.A., and Oklahoma Agricultural Experiment Station*

In the fall of 1956 numerous black mummies of the greenbug (*Toxoptera graminum* (Rond.)) were noted on wheat plants being screened for greenbug resistance in the small grain greenhouse at Stillwater, Oklahoma. Specimens sent to the United States National Museum were identified as *Aphelinus nigritus* How.[2] This species was first described by Howard in 1908.

According to Webster & Phillips (1912), this parasite has been found in Kansas, Minnesota, New Mexico, and South Carolina. Subsequent collections have been made in Arizona and California. Its hosts were listed as the rusty plum aphid (*Hysteroneura setariae* (Thos.)), *Pseudococcus* sp., and *Toxoptera* sp.

In an attempt to establish new host and temperature ranges, *nigritus* was caged with the more important aphids that attack small grains and other crops. Replicated cages containing the following species were used in several experiments at various temperature levels: English grain aphid (*Macrosiphum granarium* (Kby.)), apple grain aphid (*Rhopalosiphum fitchii* (Sand.)), corn leaf aphid (*R. maidis* (Fitch)), greenbug (*Toxoptera graminum* (Rond.)), cabbage aphid (*Brevicoryne brassicae* (L.)), and spotted alfalfa aphid (*Therioaphis maculata* (Buckton)), (also called Pterocallidium sp).

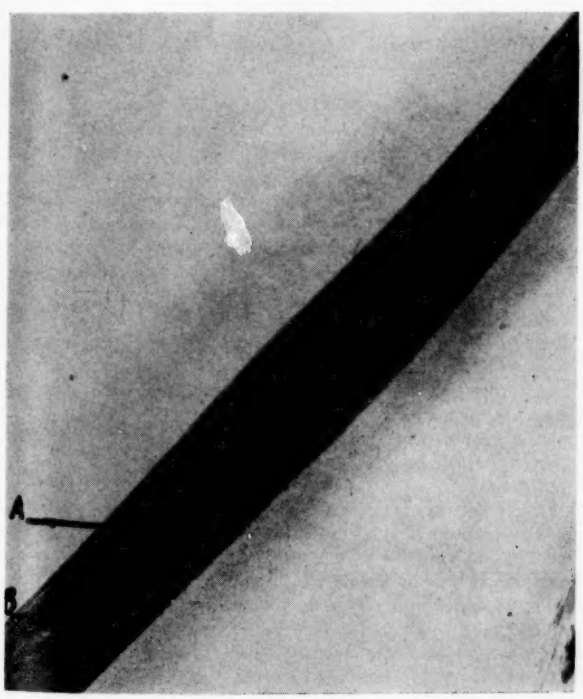


FIG. 1.—Mummified greenbugs attached to wheat leaf showing (A) *Aphelinus nigritus* How., and (B) *Aphidius testaceipes* (Cress.).

Nearly 100% parasitization was noted in cages containing the greenbug, the apple grain aphid, and the corn leaf aphid. Approximately 10% of the English grain aphids were parasitized, but the cabbage and spotted alfalfa aphids sustained no parasitization in any of the tests. The parasite was also released in the field, but no recoveries were made, possibly owing to the small populations of host material existing at that time.

Prior to the appearance of *nigritus*, the most efficient parasite of the greenbug in Oklahoma was considered to be *Aphidius testaceipes* (Cress.). Parasitization by this species is restricted to the later instars and only when temperatures are above 56° F. By comparison it was observed that *nigritus* parasitized all stages of its hosts at temperatures as low as 42°. Host material parasitized by *nigritus* becomes jet black within a few days, whereas host mummies of *testaceipes* become light golden in color (fig. 1).

REFERENCES CITED

Howard, L. O. 1908. Upon the aphis-feeding species of *Aphelinus*. Ent. News **19:** 366.

Webster, F. M., and W. J. Phillips. 1912. The spring grain aphis or "greenbug." U. S. Dept. Agric. Bull. **110:** 123.

[1] Accepted for publication April 16, 1958.

[2] Determined by B. D. Burks, of the Entomology Research Division, U.S.D.A

Tests Comparing Eight Insecticides for Control of the Mexican Bean Beetle[1]

CHARLES H. BRETT, *North Carolina State College*

and

ROSS W. BRUBAKER, *Entomology Research Division, Agr. Res. Div., U.S.D.A.*

By 1951 it had become apparent that rotenone no longer was giving satisfactory control of the Mexican been beetle, *Epilachna varivestis* Muls., in the Mountain area of Fletcher, North Carolina. Since that time tests for the control of this insect have been conducted by the North Carolina Agricultural Experiment Station and the U. S. Department of Agriculture, the latter in cooperation with the Virginia Truck Experiment Station at Norfolk. These tests established the fact that the beetle possessed a definite resistance to this insecticide (Brett & Brubaker 1955). This is still evident from tests conducted during 1956 and 1957 at Fletcher and also at Norfolk.

Many materials have been tested in an effort to find more effective toxicants, especially those with relatively low mammalian toxicity. Malathion proved to be a very desirable insecticide (Brett & Brubaker 1953) and has been widely and successfully used. Continued work has shown several other compounds offering much promise as Mexican bean beetle insecticides.

Field experiments were conducted on plots 4 rows wide and 37½ feet long. Each treatment was replicated four times. Insecticide dusts were applied with double-outlet hand-cranked dusters at the rate of 25 to 30 pounds per acre. Treatments were evaluated on the basis of populations surviving and/or visual estimates of defoliation of plants. Usually the counts were based on the number of insects on 20 plants from each of the two center rows. Data on defoliation were also obtained from the two center rows. "Per cent control" was based on comparison of the insects or de-

[1] Accepted for publication April 16, 1958.

Table 1.—Per cent control of the Mexican bean beetle obtained in field tests with insecticide dusts at Fletcher, N. C., and Norfolk, Va., 1956.

Insecticide (Per Cent of Active Ingredient)		Population Counts (Fletcher)	Defoliation Estimates (Norfolk)
Chlorthion	3	95	52
Hercules AC-528	5	91	64
Dipterex	5	90	53
Malathion	4	97	65
Rotenone	1	45	79
Sevin	2	99	95
Coefficient of variation (%)		60	67

foliation on treated plots with those on the untreated check plots. Because of the variability pattern in count data, a square-root transformation was made of the individual counts before statistical analysis. From the analysis of variance the differences required for significance at the 5% level were computed.

Data presented in table 1 show that control with rotenone at Fletcher in 1956 was less than with any of the other materials. At Norfolk, Dipterex and Chlorthion were less effective than the other materials tested.

In table 2 the data show that in 1957 rotenone was less effective than any of the other materials against third and fourth instar larvae at Fletcher. Sevin (1-naphthyl-N-methyl carbamate) and Thiodan were better than the other materials when all larvae and pupae were counted. At Norfolk there was no significant difference between any of the materials in 1957.

Many of these insecticides were also tested in the laboratory. In each test 25 adult beetles collected at Fletcher were placed in a 2-quart Mason jar and treated with a single 40-mg. puff of dust. Ten minutes later the insects were removed and placed in another jar with untreated fresh bean plants. The temperature of the laboratory was 85° F. during these tests.

In table 3 the data show that rotenone was significantly less effective than any of the other materials after 4, 8, and 42 hours. Hercules AC-528 (Delnav) was significantly less effective after 4 and 8 hours but not after 24 hours.

It is apparent from the 1956 and 1957 data that rotenone continued to be ineffective against the Mexican bean beetle at Fletcher. Hercules AC-528 was a slow-acting insecticide, but after 24 hours it gave control comparable to other materials tested. Chlorthion and Dipterex gave good control at Fletcher, but were less effective than the other materials at Norfolk. Diazinon and malathion gave good control at both places, but at Fletcher they were less effective than Sevin or Thiodan when all

Table 2.—Per cent control of the Mexican bean beetle obtained in field tests with insecticide dusts at Fletcher, N. C., and Norfolk, Va., 1957.

Insecticide (Per Cent of Active Ingredient)		Fletcher, N.C. (Population, Counts)		Norfolk, Va.	
		Third and Fourth Instars	All Larvae	Population Counts[a]	Defoliation Estimates
Diazinon	4	90	77	98	84
Malathion	4	85	73	97	85
Rotenone	1	0	50	95	94
Sevin	1	85	99	100	85
	2	92	99	100	95
Thiodan	4	44	97	95	93
Coefficient of variation (%)		58	34	48	146

[a] Third and fourth instar and pupae.

Table 3.—Per cent mortality of Mexican bean beetle adults at various intervals after treatment with different insecticides, 1956.

Insecticide (Per Cent of Active Ingredient)		Per Cent Mortality After Hours Indicated		
		4	8	24
Chlorthion	3	68	86	90
Hercules AC-528	5	6	42	74
Dipterex	5	94	96	98
Malathion	4	92	92	98
Rotenone	1	18	24	42
Sevin	2	60	74	92
No treatment		0	0	0
Coefficient of variation (%)		13	8	9

larvae and pupae were counted. Thiodan appeared to be less effective than the other materials at Fletcher when counts were made of third and fourth instar larvae, but this difference was not significant. Sevin was consistently very effective at 1% or 2%. The 2% dust showed only slight superiority. With the relatively low mammalian toxicity reported for this insecticide and its consistent excellence, it should become one of the most useful materials for Mexican been beetle control.

References Cited

Brett, Charles H., and Ross W. Brubaker. 1953. Mexican bean beetle control with malathion compared with eight other materials. Jour. Econ. Ent. **46**(5): 912–3.

Brett, Charles H., and Ross W. Brubaker. 1955. Rotenone resistance in the Mexican bean beetle. Jour. Econ. Ent. **48**(3): 343.

Control of the Clover Root Curculio on Alsike Clover[1]

E. A. Dickason, C. M. Leach, and A. E. Gross,[2]
Oregon State College, Corvallis

There are many published accounts of the clover root curculio, *Sitona hispidula* (Fabr.), injuring legume and grass crops. Underhill *et al.* (1955) and Turner (1957) controlled the clover root curculio on alfalfa in Virginia by incorporating insecticides in the soil prior to planting, and by applying insecticides on the soil surface in spring or fall. In general, best results were obtained from applications of chlorinated hydrocarbon insecticides such as dieldrin, aldrin, heptachlor, or chlordane.

In Oregon, the clover root curculio is believed to be a major cause for the decline of alsike clover (*Trifolium hybridum*) seed yields because of its effect on plant vigor and stand. Larvae damage the root system by feeding on root nodules, small secondary fibrous roots, and by gnawing deep cavities in the tap root. In addition to the mechanical injury caused by larvae, there is also a possible relationship between insect damage and fungus root-rots. Leaf injury by adults in early spring is of minor importance.

Early spring application of DDT (1 lb. per acre) in Oregon controls adults and reduces root injury. Frequently high percentages of roots are injured either by larvae hatching from eggs laid during periods of mild weather in fall, winter, or early spring, or by larvae hatching from eggs deposited in late spring by adults migrating into treated fields. Soil insecticide plots were established in 1955 in an attempt to control larvae as they moved through the soil to the root area of the plant. Control would per-

[1] Approved for publication as technical paper, number 1085, Oregon Agricultural Experiment Station. Accepted for publication April 21, 1958.

[2] Assistant Entomologist, Assistant Plant Pathologist, and Superintendent, Klamath Branch Experiment Station, respectively.

Table 1.—Control of the clover root curculio with heptachlor applied to the soil at the rate of 4 pounds per acre in the spring of 1955. Klamath Falls, Oregon.

Treatment	Insect		Vascular Rot		Cortical Rot	
	Injury Index[a]	Per Cent Damaged	Injury Index[a]	Per Cent Damaged	Injury Index[a]	Per Cent Damaged
			1956			
Untreated		86				
Heptachlor		3				
			1957			
Untreated	2.4	98	0.4	16	0.2	12
Heptachlor	0.6	38	0.3	10	0.1	2

[a] Average index, injury rated: 0, none; 1, slight; 2, moderate; 3, severe; and 4, plant dead.

mit an evaluation of possible relationships between root injury and fungus root-rots.

In the spring of 1955 a field at Klamath Falls, Oregon, was divided into 32- by 180-foot plots. Emulsifiable concentrate heptachlor was applied at the rate of 4 pounds of actual toxicant per acre to three of the plots and disked into the soil. Three plots were disked and left as untreated checks. The field was seeded to oats following treatment and alsike clover was drilled in the oat stubble in late summer.

Clover seed yields were not taken because of lack of uniformity in stand caused by alkali areas in the soil and by severe mouse damage during the concluding year of the experiment. Control was evaluated by scoring roots for insect injury in the fall of 1956, and for insect injury and disease incidence in the fall of 1957. The results are summarized in table 1.

As indicated in the 1956 counts (table 1), heptachlor gave excellent protection to the roots throughout the first seed crop year. In 1957 (table 1) there were more plants attacked in treated plots than in 1956, suggesting that heptachlor had lost some of its effectiveness in controlling larvae. However, as indicated in table 1, the injury index average for the treated plants was only 0.6 in contrast to 2.4 for plants in the untreated checks. The high insect injury index in the check plots is a result of the additive effect of 2 years injury.

There were no marked differences in the incidence or severity of root-rots of two-year-old plants sampled from the heptachlor and check plots. However, there was a low incidence of root-rots throughout the plot area.

It is unfortunate that seed yields could not be taken to correlate the possible benefits of insect control with seed production. The use of soil insecticides appears to be a very promising method of reducing clover root curculio injury to alsike clover roots. Whether the prevention of root damage will lessen the invasion of roots by fungus pathogens has yet to be shown.

References Cited

Turner, E. C., Jr. 1957. Control of the clover root curculio in alfalfa. Jour. Econ. Ent. 50(5): 645–8.

Underhill, G. W., E. C. Turner, Jr. and R. G. Henderson. 1955. Control of the clover root curculio on alfalfa with notes on life history and habits. Jour. Econ. Ent. 49(2): 184–7.

Economic Importance and Control of the Loblolly Mealybug, *Dysmicoccus obesus* Lob.[1, 2]

H. E. Milliron, *Glen Dale, West Virginia*

Late in July, 1956, a heavy mealybug infestation was discovered in an ornamental planting of 15-year-old loblolly pines, *Pinus taeda* L. in New Castle County, Delaware. Mature specimens of the pest were collected and identified as *Dysmicoccus obesus* Lobdell, which was originally described from Mississippi,

Fig. 1.—Ornamental loblolly pine, showing typical effects of mealybug infestation.

and heretofore apparently never reported as being destructive anywhere. The source of the trees in the above planting was not established, and subsequent extensive search in several natural stands of this pine throughout the southern half of the state did not reveal other infestations.

The insect was present on 12 of the 36 trees, and was entirely confined to the main trunk from ground level to a maximum height of 4 feet. From a considerable distance, the infested trees, of which a typical example is shown in figure 1, were readily distinguished from non-infested ones by their spindly appearance, sparser foliage, shorter height and off-color. The most severely infested tree had the basal portion of its trunk enlarged and was, in this respect, out of proportion with what is regarded as the central leader. A conspicuous, white powdery deposit in cracks and crevices of the bark, as illustrated in figure 2, indicated the occurrence of mealybugs, which were readily found beneath the bark scales on the outer cambium. Many of the larger specimens were located in any suitable cavities beneath the bark.

Observations were continued through the summer of 1956, and into the fall. At least two species of ants, one of which was *Lasius alienus americanus* Emery, were common on and about trees harboring the mealybug. Only occasionally was an ant observed on a normal tree.

Additional mealybug collections were made in mid-March, 1957. Numerous first instars were found active and feeding in well-protected places beneath the bark scales. No viable specimens larger than the first instar were located at that time, indi-

[1] Hemiptera: Pseudococcidae.

[2] Accepted for publication April 21, 1958.

FIG. 2.—Trunk of loblolly pine infested with mealybugs. Note white powdery deposit in cracks and crevices of bark.

cating that the species overwintered above ground in either that stage or as eggs.

CONTROL.—A single application of malathion (55% emulsifiable concentrate) at the rate of 2 tablespoons per gallon of water per tree, was made on March 31, 1957, using a minimum pressure of 50 p.s.i. The weather was bright and clear, with seasonable temperature and little wind. Although entire trees were treated, special attention was directed to the lower portions of the trunk. A careful check of several trees, made 1 week later, disclosed that the treatment apparently was 100% effective. Moreover, no living material was found during any of several examinations conducted between that time and May 15, when control was begun against the Nantucket pine moth, *Rhyacionia frustrana* (Comst.). Evidently, the mealybug was completely eliminated from that planting by the single, thorough application of malathion in late March. No phytotoxic effects were noted under these conditions.

Additional Host Records for Two Bostrichid Powder-Post Beetles[1]

RICHARD H. SMITH, *California Forest and Range Experiment Station, Forest Service, U. S. Department of Agriculture*

The lead cable borer, *Scobicia declivis* (Lec.), is a native pest which has been recorded as a borer in several species of seasoned hardwoods. The beetle has been reported most often in California where it apparently maintains itself on cut or dead wood. In the past it has shown some unique habits. One of these, boring into lead coating used to sheath telephone cables, is the basis for its common name. At times it has been recorded boring into wooden wine casks and asphalt roofing. Its habits have been fairly well described (Burke *et al.* 1922).

In 1956 the beetles were found attacking Philippine mahogany lumber that was piled for seasoning at a California sawmill. The species of mahogany could not be determined with certainty because no effort had been made to keep logs and lumber separated during the various handling processes. The best guess is that the wood consisted primarily of the genus *Shorea*. This mahogany had been imported as logs from the Philippines and sawed into lumber at a mill in El Dorado County.

The attacks took place in the spring; no attacks were noted in mid- or late summer. An examination of the lumber showed that the attacks were confined to the sapwood and were characterized by a circular hole 1.5 to 2.5 mm. in diameter. The gallery went straight into the wood for one-fourth inch or more, then often made a right angle turn and continued parallel to the grain of the wood. As far as could be determined, these habits are similar to those on its natural hosts.

A small sample indicated one or more holes in about 15% of the exterior boards in several of the stacks of seasoning lumber. The average was about 6 holes; the range, 1 to 15. This damage was not yet serious since the lumber was largely heartwood which the beetle apparently did not attack.

Two facts, however, pointed to the necessity for some kind of action. First, the Sierra foothill country in the vicinity of the mill could be a continuing source of beetle population. Second, though a cursory examination indicated no regeneration of the beetle in the wood, there was no positive assurance that the beetle could not multiply in mahogany. Therefore, the stacks of lumber were sprayed with an aqueous emulsion containing 0.25% gamma isomer of BHC. A subsequent examination revealed no brood development and no new attacks. Either the spray was effective, the beetle was unable to reproduce in mahogany, or there were no beetles in the area.

This incident is somewhat similar, though more or less reversed, to that of *Heterobostrychus aequalis* (Waterh.) in hickory, *Carya* sp. In 1949, this insect was found in the hickory sections of a great number of Army cots brought back to the United States from the Philippines. This large bostrichid, a native to some of the islands of the Pacific, is a pest of considerable importance in mahogany in the Philippines, and it has been frequently found in foreign wood products imported to the United States (Fisher 1950, St. George 1948). However, in the case cited here, it readily attacked the sapwood of hickory when that wood was in the Philippines. Both living larvae and adults were found in the wood when the cots were returned to the United States. Even though it was doubtful that the insects could regenerate in the wood in this country, the cots were immediately fumigated.

Thus we have an illustration of two beetles readily attacking woods foreign to their habitat. This experience points to the need for continued surveillance of the habits of our native pests and the habits of foreign pests on material native to this country.

REFERENCES CITED

Burke, H. E., R. D. Hartman, and T. E. Snyder. 1922 (Rev. 1923) The lead cable borer or "short-circuit beetle" in California. U. S. Dept. Agric. Bull. **1107.** 56 pp., illus.

Fisher, W. S. 1950. A revision of the North American species of beetles belonging to the family Bostrichidae. U. S. Dept. Agric. Misc. Publ. **698.** 157 pp. (p. 86).

St. George, R. A. 1948. Remarks at meeting. Ent. Soc. Wash. Proc. **50**(5): 138.

[1] Accepted for publication April 25, 1958.

An Egg Survey of *Hippelates* Gnats in the Vicinity of Orlando, Florida[1]

MARTIN L. TAYLOR and L. D. OLINGER, *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*

Hippelates gnats (Diptera, Chloropidae) are annoying pests of man and domestic animals. Because they feed at the eyes and other natural orifices, they probably aid in the spread of diseases.

During the summer of 1956 a method for conducting a *Hippelates* egg survey was developed and applied to four types of potential breeding areas in the vicinity of Orlando, Fla. The principal species involved were *pusio* Loew and *bishoppi* Sabr., but no attempt was made to separate the eggs by species.

A soil sampler, consisting of a copper pipe 8 inches long and 4 inches in diameter, was driven several inches into the ground. The top half-inch of soil was removed with a spoon, and placed in a labeled pint jar. Usually three samples were placed in each jar.

In the laboratory samples from each jar were screened into a pan, and mixed with water containing magnesium sulfate (3 to 1 by volume) to increase buoyancy of the eggs. The mixture was thoroughly stirred and allowed to settle for 1 to 2 minutes. The supernatant water was decanted through a sieve with 0.044-mm. openings. The eggs and debris that accumulated in the sieve were washed to remove the scum, and then transferred into another pan containing water and magnesium sulfate. The liquid phase was poured into pint jars, and the eggs were counted under a binocular microscope.

Four types of areas were surveyed—vegetable fields, citrus groves, flower beds around residences or parks, and lake margins. Results of the survey are shown in table 1.

The number of *Hippelates* eggs collected in the various samples ranged from 0 to 75, but was usually between two and four. The highest density was found in vegetable fields, followed by citrus groves and flower beds.

The prevalence of *Hippelates* eggs in flower beds may account for the large adult populations sometimes found in residential sections where larger cultivated areas are absent. Eggs were not found along lake margins, although large populations of adult gnats were present during the summer. Moisture content apparently was an important factor, for soils containing 5% to 15% were most favorable for oviposition.

Eggs were found only in comparatively loose soil. In vegetable fields the peak in oviposition occurred between the second and eighth days after cultivation. While cultivation has an important effect on soil looseness, some soils remain loose for longer periods than others. The heavier types of soil, such as clay and silt, may pack a few days after they have become moistened by rain and become unsuitable for breeding until they are cultivated again. Sandy soils, however, remain loose for several weeks or months after cultivation. Eggs were found in vegetable fields that had remained uncultivated for 3 months where the soil was protected from rain by cover crops. They were also collected in loose soil in citrus groves that were covered with grass 12 to 15 inches tall.

Eggs were collected from soil ranging in pH from 5.8 to 7.6. The numbers of eggs collected per square foot during July, August, and September 1956 are shown in table 2. During July the method for separating the eggs from the soil was being developed, and full proficiency in detecting their presence was not obtained. This may account for the lower count during that month than during August and September, when they were nearly equal.

Table 1.—Results of a *Hippelates* egg survey in four types of areas near Orlando, Fla.

Type of Area	Number of Samples	Number of Eggs per Square Foot	
		Hatched	Total
Vegetable fields	71	6.8	12.9
Citrus groves	121	5.6	8.7
Flower beds	71	4.7	6.8
Lake margins	48	0	0

Table 2.—Number of *Hippelates* eggs per square foot collected in the summer of 1956.

Month	Hatched	Total
July	4.4	6.6
August	7.3	11.0
September	7.5	11.1

Single tests were conducted in a citrus grove and vegetable-growing area to determine the time of day oviposition occurred. In each area the top 3 inches of soil was removed from a plot 8 feet square to remove any eggs that might be present. The plots were spaded to thoroughly loosen the sod, and four soil covers 3 feet square and 8 inches high were placed on each of them. The covers were removed and replaced one at a time to expose portions of the soil to oviposition for 6 hours. To aid in attracting adult gnats to the areas, fish heads were placed in three pint jars with hardware-cloth caps and kept in portions of the plots that were uncovered. The test conducted in the citrus grove lasted 4 days, and each cover was removed four times for a total of 24 hours. The test in the vegetable area lasted 3 days, and each plot was exposed for 18 hours.

All ovipositions occurred during daylight, for no eggs were deposited between 9 P.M. and 3 A.M. The largest numbers, ten in the citrus grove and six in the vegetable area, were deposited between 9 A.M. and 3 P.M. There were about twice as many daylight hours during this interval as during any of the other exposure periods. Between 3 and 9 P.M. another three were found in the citrus grove and four were present in the vegetable area. The only two laid between 3 and 9 A.M. were in the vegetable area.

[1] Accepted for publication April 28, 1958.

Trunk Sprays for Control of the Peach Tree Borer[1]

OLIVER I. SNAPP, *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*[2]

Trunk sprays of DDT and BHC tested at Fort Valley, Georgia, during the period 1945–51 showed promise for control of the peach tree borer (*Sanninoidea exitiosa* (Say)), especially where there were less than two live borers per tree in the untreated trees (Snapp 1950, 1952).

Experiments with DDT and BHC were continued in 1951–56, and parathion was also included. All sprays were applied with a power sprayer to the trunks and lower part of large limbs. Four applications were made at about monthly intervals beginning about August 1, except in 1955, when three applications were made beginning July 15 to the 2- and 3-year-old trees. There was no preparation of the soil or other attention given the trees except for removal of interfering high grass and weeds. Ethylene dichloride emulsion was used in each experiment as a basis of comparison. It was poured around the base of the tree about October 23 in such a way that the soil absorbed and held it at the ground line. The soil level was raised before this treatment where there were signs of borers in the tree above the normal level. A little soil was placed on the treated surface to prevent surface loss of the fumigant. Each treatment was replicated five times

[1] Accepted for publication April 28, 1958. Presented at the meeting of the Entomological Society of America at Memphis, Tenn., December 2–5, 1957.

[2] Julius P. Hollon assisted with these experiments.

Table 1.—Numbers of live peach tree borers per tree after treatment with trunk sprays or ethylene dichloride soil applications, 1951–56.

Treatment (Quantity per 100 Gallons or per Tree)	Age of Trees (Years)				
	2	3	4	5	6–8[a]
1951–52					
Parathion (15%) 2 lb.	—	—	—	0.4	0.2–0.6
BHC (6% gamma) 8 lb.	—	—	—	.6	0.1–0.2
Ethylene dichloride (20%) 0.5 pt.	—	—	—	0.3	0.1–0.1
Untreated check	—	—	—	3.8	2.4–5.4
1952–53					
Parathion (15%) 2 lb.	0	—	—	0.8	—
DDT (75%) 5.3 lb.	0	—	—	0.7	—
BHC (10% gamma) 4.8 lb.	0	—	—	0.1	—
Ethylene dichloride (15%) 0.25 pt.	0	—	—	—	—
(20%) 0.5 pt.	—	—	—	0.3	—
Untreated check	0.4	—	—	7.7	—
1953–54					
Parathion (15%) 2 lb.	0	2.2	0.9	—	0
DDT (75%) 5.3 lb.	0	1.0	0.5	—	0
BHC (10% gamma) 4.8 lb.	0	1.0	0.5	—	0
Ethylene dichloride (15%) 0.25 pt.	0	—	—	—	—
(15%) 0.5 pt.	—	0.1	—	—	—
(20%) 0.5 pt.	—	—	0.1	—	0
Untreated check	0.6	6.8	4.9	—	0.2
1954–55					
Parathion (15%) 2 lb.	0	0	1.4	0.5	—
DDT (75%) 5.3 lb.	0	0	1.1	0.8	—
BHC (10% gamma) 4.8 lb.	0	0	0.6	0.2	—
Ethylene dichloride (15%) 0.25 pt.	0	—	—	—	—
0.5 pt.	—	0	—	—	—
(20%) 0.5 pt	—	—	0.6	0.2	—
Untreated check	0.6	1.6	5.7	2.3	—
1955–56					
Parathion (15%) 2 lb.	0	0.3	—	2.2	1.5
DDT (50%) 8 lb.	0	0	—	4.4	0.2
Ethylene dichloride (20%) 0.5 pt.	—	—	—	0.2	0.1
Untreated check	1.3	3.6	—	12.9	5.1

[a] 7–8 years in 1951, 6 years thereafter.

in randomized plots of five trees each. All trees were examined in the following spring for borer control and tree injury.

The results are given in table 1. The trunk sprays were highly effective against light infestations but were not dependable against moderate to heavy infestations. Five borers per tree of prime production age was considered a moderate and more than eight a heavy infestation. This result is believed to be due to the long oviposition period of the insect in this latitude. Moths have been observed in peach orchards as early as April 25, and eggs have been known to hatch as late as December 7. The moths usually begin to deposit eggs on the day of emergence. In heavily infested peach orchards the spread of the usual three or four trunk sprays per season apparently is not sufficient to insure a high degree of control. Ethylene dichloride emulsion as applied once in the fall gave very good control of the peach tree borer regardless of the degree of infestation.

References Cited

Snapp, Oliver I. 1950. DDT and some of the newer insecticides for control of peach tree borers. Jour. Econ. Ent. **43**(3): 315–8.

Snapp, Oliver I. 1952. Benzene hexachloride and DDT sprays for peach tree borer control. Jour. Econ. Ent. **45**(3): 547.

The Effects of Dow ET-57[1] on Chicken Shaft Louse Infestations[2]

E. M. Raffensperger, *Virginia Agricultural Experiment Station, Blacksburg*

The search for an insecticide systemic in action against parasites of warm-blooded animals received a strong push forward in 1955, at which time it was announced by letter from the United States Department of Agriculture that the material now known as Dow ET-57 showed promise of cattle grub control. More recently, work conferences[3] on the use of this material and other systemic insecticides have been conducted with enthusiasm, and tests against parasites of sheep and swine have been added to the earlier work with cattle. The use of Dow ET-57 in poultry parasite control studies has been neglected by many workers because the hazards of toxic residues in poultry products were considered too great.

In 1956 and 1957 chickens from the Virginia Agricultural Experiment Station flock were used in testing Dow ET-57 against populations of the shaft louse of chickens, *Menopon gallinae* (L.). A pilot test indicated that doses of approximately 250 milligrams of Dow ET-57 per kilogram of body weight of the infested bird would be needed for louse control. Chicken eggs, collected during the pilot test and incubated under commercial hatchery conditions, indicated no interference with hatching from the insecticide at this dosage. Dosages beginning with 250 milligrams of the toxicant per kilogram of body weight and running to 1000 milligrams per kilogram, as shown in table 1, were then administered to groups of 10 chickens. The birds were mature White Plymouth Rocks of uniform size and condition, selected from a laying flock, and all were heavily infested with lice. The insecticide, in the form of a 50% emulsifiable concentrate, prepared for veterinary use, was administered orally by means of a calibrated syringe and a catheter tube. A four-point scale was used for scoring louse populations at frequent intervals following treatment.

One untreated male chicken with a severe infestation of lice was maintained with each treated group to provide a source of lice for reinfestation.

On the fourth day following treatment a single treated bird, given 250 milligrams of the toxicant per kilogram of weight, was found to have living lice. No lice were found on any other treated bird. A group of 10 untreated birds showed no reduction in louse population.

All treated birds were examined daily for gross symptoms of toxicity. At the top level of treatment, 1000 milligrams per kilogram, two birds exhibited symptoms of typical phosphate insecticide poisoning on the fourth day following treatment. These two birds died on the fifth day. A post mortem examination showed that a complication of pneumonia was involved in the deaths.

During the latter part of the 30-day test, birds treated with

[1] Pharmaceutical grade; also called Trolene.

[2] Accepted for publication May 2, 1958.

[3] Kerrville, Texas, September 15 and 16, 1956; Kerrville, Texas, July 9 and 10, 1957.

Table 1.—Indices[a] of louse infestations for groups of 10 birds following single oral doses of Dow ET-57.

Dosage (mg./kg.)	Days Following Treatment												
	0	1	2	3	4	6	8	10	13	16	21	26	30
250	29	3	2	2	1	2	3	6	8	9	14	14	18
500	29	3	0	0	0	0	0	3	5	7	19	17	20
750	29	1	0	0	0	0	0	0	3	5	7	6	7
1000[b]	30	6	4	2	0	0	1	0	3	2	5	6	6

[a] Each of 10 birds was rated on an arbitrary scale from 0, no lice, to 3, a heavy infestation. Each index is a sum of 10 ratings.

[b] Corrected to nearest whole number for 10 birds after the death of two birds on the fifth day following treatment.

Table 2.—Pounds of Dow ET-57-treated feed refused by groups of 27 chickens in a 6-day feeding test.[a]

Dow ET-57 in Feed (p.p.m.)	Pounds Refused in Consecutive 2-Day Periods			Total Refused
	1–2	3–4	5–6	
0	0	0	0	0
100	0	0	0	0
200	1.7	0	0	1.7
300	4.4	0.2	2.0	6.6
400	3.9	1.1	3.2	8.2
500	6.8	3.6	4.3	14.7

[a] Feed was mixed at 2-day intervals, at which times refused feed was removed from pens and weighed.

250 or 500 milligrams per kilogram exhibited rapidly increasing louse populations, as shown in table 1, while birds receiving 750 or 1000 milligrams per kilogram acquired only small numbers of lice.

In a search for an easier means of administering the toxicant, a test of palatability of feed-insecticide mixtures was conducted. Dow ET-57 as a 25% wettable powder was incorporated into a standard laying mash at rates of 100 to 500 parts of the toxicant per million of feed. Twenty-seven birds were fed a treated ration *ad lib.* at each level. Table 2 shows total feed refused by each group during the six-day test, the highest level of treatment showing the greatest amount of food refused. Such a drop in food consumption in a commercial flock would lead to a prohibitive loss of production. Although all birds were heavily infested with lice at the beginning of the palatability test, no increase in numbers of lice was revealed in daily scorings of the populations, even at the highest dosages. A calculation of toxicant consumed on a body weight basis indicated that feeding dropped off rapidly before a level of 25 milligrams of the insecticide per kilogram of body weight per day was reached.

The Effect of Residual Barn Sprays on Control of Horn Flies[1]

E. C. TURNER, JR., AND L. T. HARGETT,
Virginia Agricultural Experiment Station, Blacksburg

Recent work by several investigators has shown that the application of Diazinon to the walls, ceiling and stanchions of dairy barns as a residual spray for the control of house flies (*Musca domestica* L.) also has resulted in a reduction in the number of horn flies (*Siphona irritans* (L.)) on cows which were being brought into these barns for milking and feeding. Hansens (1956), in New Jersey first noted this effect and reported that horn flies were reduced on dairy cows being brought into these barns, sprayed with Diazinon, for as long as 2 months. He concluded that the flies were killed in part by fumigation and in part by contact with this material. That same year, Goodwin (1956) demonstrated that horn flies, exposed for 2 hours to the vapors of Diazinon sprayed as 0.5% suspension of wettable powder 25% Diazinon, resulted in 100% kill of horn flies for 6 days after spraying.

These results seemed to indicate that residual applications of Diazinon, used primarily to control house flies, would also constitute an effective control of horn flies and thereby would eliminate, at least in part, direct treatment of dairy animals. Thus it was proposed to determine if the reduction in horn fly population by these residual sprays would constitute an effective control, and if other insecticides, applied primarily to control house flies, would also control horn flies.

PROCEDURE.—In the summer of 1956, preliminary tests were established in combination with house fly control experiments. Several barns were sprayed, by means of a high pressure sprayer,

Table 1.—Preliminary results showing the effect of residual sprays on control of horn flies. Montgomery County, Va. 1956.

Insecticide and Per Cent Concentration[a]	No. Treatments	Range in No. of Weeks Satisfactory Control[b]
Diazinon, 1	4	2 to 7
Dow ET-57	6	1
Lindane, 0.3	3	2 to 3

[a] Wettable powder 25% formulations were used to make up all treatments.
[b] At least 60% reduction of horn flies from the pretreatment counts.

with solutions of 1% Diazinon, 1% Dow ET-57[2] and 0.3% lindane. The wettable powder 25% formulation was used with all three of these materials. All animals in these barns were milked twice a day and were held in the barns for approximately the same length of time. Horn fly counts were made in the pastures once a week on 10 cows randomly selected in each herd. When the horn fly population showed less than 60% reduction from the pretreatment counts, the control was considered unsatisfactory.

In 1957, another experiment was designed to test the effect on horn fly population of several other insecticides that were currently being tested for control of house flies in dairy barns. It was desired, in addition, to determine what effect ventilation in the barns might have on the effectiveness of these materials in controlling horn flies.

Counts of the numbers of horn flies from each herd were made in the same manner as described for the 1956 experiments. In this case, however, an untreated herd was used as a check to compare with the treated herds. Control of horn flies was considered unsatisfactory if a herd averaged more than 50 flies per animal. In the untreated herd, the population of horn flies did not go below 70 per animal at any time during the test, and most of the time the population averaged 138 per animal.

EXPERIMENTAL RESULTS.—It is noted in table 1 that the horn flies were controlled satisfactorily for at least 1 week by all materials used in the test. It is further noted that the Diaznon treatment resulted in 2 to 7 weeks satisfactory control, however the length of control was quite variable within the individual

[1] Presented at the Fifth Annual Meeting of the Entomological Society of America, held at Memphis, Tenn. Dec. 2–5, 1957. Accepted for publication May 7, 1958.
[2] Technical grade; also called Korlan.

Table 2.—The effect of barn ventilation on the use of residual sprays for control of horn flies. Montgomery County, Va. 1957.

Insecticide and Per Cent Concentration	No. Treatments	Ventilation	No. Weeks Satisfactory Control[a]
Methoxychlor, 0.5	1	Open	0
Malathion, 1	2	Open	0
	2	Closed	0
Lindane, .3	2	Open	1 to 2
	2	Closed	3
Dow ET-57, 1	2	Open	2 to 3
	2	Closed	3
Diazinon, 1	1	Open	5
	1	Closed	7
Bayer 21/199, 0.25	2	Open	1 to 3
	2	Closed	3

[a] Less than 50 horn flies per animal.

treatments. This was thought to be due to variation in the ventilation in certain barns. Some barns were completely closed and others were well ventilated when the cows were in the pastures. These practices of leaving the barns closed or well ventilated depended on the habit or preference of the dairyman.

In 1957, another experiment was designed to test this possible effect of air ventilation on the length of satisfactory horn fly control in dairy barns. Several other insecticides that were currently being tested for control of house flies were also included in the test.

The results of the 1957 experiment are shown in table 2. All treatments were duplicated except with the Diazinon and the methoxychlor. The methoxychlor and malathion treatments resulted in no satisfactory control of the horn flies. All other materials showed control in varying degrees. Ventilation of the barn seemed to have a definite influence on the period of effectiveness of the materials tested.

It can be concluded from these experiments that residual spraying of the walls, ceilings, and stanchions of dairy barns, using Diazinon, lindane, Dow ET-57, and Bayer 21/199, primarily for control of house flies can also result in a supplementary control of horn flies on dairy cows being brought into the barns for milking and feeding. It is further concluded that the period of effectiveness by this method can be increased by decreased ventilation of the barn during the time the cows are in pasture.

REFERENCES CITED

Goodwin, W. J. 1956. Toxicity of Diazinon vapors to horn flies. Jour. Econ. Ent. 49(3): 406–7.

Hansens, E. J. 1956. House fly control in dairy barns. Jour. Econ. Ent. 49 (1): 27–32.

Tests of Pathogens for the Control of Tobacco Insects[1]

F. S. CHAMBERLIN and S. R. DUTKY, *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*

Preliminary tests of several pathogens for the control of tobacco insects were carried out at Oxford, North Carolina, in 1955 and 1956. The pathogens were cultured at Beltsville, Maryland, and included the fungus *Spicaria rileyii*, a polyhedrosis virus from *Heliothis* sp., a parasitic bacterium carried by the nematode DD-136, and several spore-forming bacteria. The test insects were the tobacco budworm (*Heliothis virescens* (F.)) and the tobacco hornworm (*Protoparce sexta* (Johan.)) on flue-cured tobacco. Relatively small numbers of the corn earworm (*H. zea* (Boddie)) and the tomato hornworm (*P. quinquemaculata* (Haw.)) were also present.

INSECTARY STUDIES.—Limited studies were carried out in an insectary in which the hornworms and budworms were confined on small plants or suckers kept alive by having their stems immersed in water. Usually 5 to 10 larvae were confined on each plant and several plants used in each test. Control larvae confined in a similar manner showed only a slight mortality.

Relatively high concentrations of *Spicaria rileyii* applied to second- and third-instar hornworms topically with a camel's-hair brush and as sprays caused no mortalities under a considerable range of temperature and humidity conditions.

The polyhedrosis virus from *Heliothis* was also found to be nonvirulent against both species of hornworms.

Bacterial cultures designated as DD-133-2 and DD-133-6 caused 70 to 80% budworm mortality within 4 to 5 days, and gave 93 to 100% mortality of second- and third-instar hornworms within 3 days.

The parasitic nematode DD-136, and its associated bacterium, gave only 30% mortality of hornworms under insectary conditions when the temperature ranged from 47° to 96° F. during the test period, but killed 50% when hornworms were held in the basement at 74° to 77°. On tobacco leaves kept moist and at a nearly constant temperature of 76° the treatment gave 66% mortality within 24 hours and killed all the hornworms within 48 hours. Against budworms this treatment gave results similar to those obtained with hornworms.

FIELD TESTS.—Field experiments with pathogens were confined to the control of budworms on tobacco. A series of replicated-plot tests were carried out with the nematode-bacterium treatment, the polyhedrosis virus, the fungus *Spicaria rileyii*, and bacterial culture DD-133-2. The pathogens in water suspensions were applied to the infested plants by means of hand sprayers. Mortality records were obtained from approximately 75 budworm larvae receiving each treatment. These larvae were of all sizes, but most of them were about half grown.

In one experiment, in which the foliage was wet and the humidity high from heavy rains and the daily maximum temperature was about 87° F., the nematode, applied at a rate of 5 million per 100 plants, caused a larval reduction of 80 to 85% within 3 to 4 days, but there was little further reduction. In other experiments, in which the temperature frequently exceeded 90° and the surface of the tobacco leaves was dry during much of the test period, the nematode gave little or no control.

An application of the virus at 48.75 billion polyhedra per 100 plants showed evidence of a slow effectiveness against budworm larvae under a wide range of temperature and moisture conditions and caused 100% mortality in 13 days. Ten days were required for an adequate reduction, during which period the tobacco plants sustained severe feeding damage. Although the virus failed to control budworms within the 2- to 4-day period usually considered essential by tobacco growers, its action might be considered satisfactory on crops having less exacting cultural demands.

The fungus *Spicaria rileyii*, applied at the rate of 1500 billion spores per 100 plants, caused a slight budworm mortality but failed to give adequate control under a considerable range of climatic conditions.

The bacterial disease, applied at the rate of 94 billion spores per 100 plants, reduced budworm populations in low varying amounts, but not sufficiently to meet the cultural demands.

[1] In cooperation with North Carolina State Department of Agriculture and North Carolina State College. Accepted for publication May 12, 1958.

Toxicity of BHC in Milk to House Flies as Related to Butter Fat Content of the Milk[1]

FRANK W. FISK, *Department of Zoology and Entomology, The Ohio State University, Columbus*

As part of a controlled feeding and residue study two groups of lactating dairy cows, Holstein and Jersey-Guernsey, were stanchioned and restricted to diets to which sufficient amounts of technical BHC (40% gamma) were added to provide the following concentrations of total BHC in p.p.m. of diet: 0, 1, 5, 25, and 125.[2] After 50 days on this restricted diet the cows were allowed on pasture and no more BHC was given. Milk samples were withdrawn at weekly intervals during the 50-day feeding period and thereafter so long as available and positive for BHC. The chemical analyses, a modified Schechter-Hornstein method, for total BHC were run on all samples and are reported elsewhere (Ware 1958). House fly bioassays for BHC were run on many of the samples. The whole-milk feeding technique described by Sun & Sun (1953) was followed.

Standard concentrations of 0.5 to 2.0 p.p.m. lindane in milk were prepared, using milk from the check (0 p.p.m.) cows and

[1] Department of Entomology, Ohio Agricultural Experiment Station Journal Article No. 41-58—U.S.D.A. Cooperative Regional Project NC-33, with the cooperation of the Dairy Science Department, Ohio Agricultural Experiment Station, Wooster. Accepted for publication May 15, 1958.

[2] The actual p.p.m. of tech. BHC fed test animals based on daily feeding records was: 1, 0.88; 5, 4.00; 25, 21.77; 125, 102.13.

adding the lindane in acetone. Samples from the BHC-fed cows, except the maximum 125 p.p.m. rate, were fortified with 0.8 p.p.m. lindane before test. All samples were heated to 65° to 70° C. and homogeneized by hand or with a Waring blender before fortifying and testing. Replicated 20-ml. aliquots of the samples were poured onto cellucotton in 1-oz. paper souffle cups and these placed in wide-mouth pint Mason jars with fresh cheesecloth covers. Under CO_2 anesthesia 50 unsexed adult house flies, 18 to 24 hours after emergence, were counted into each jar. Mortality was recorded after 48 hours incubation at 25° C. Concentrations of BHC in the samples were estimated by plotting mortalities of the unknowns on the regression line derived from the lindane standards for that day.

Calculations from the bioassay data of total BHC excreted in the milk (*e.g.* gamma BHC×2.5) reached a maximum of 5 p.p.m. in the milk of cows fed 125 p.p.m. BHC, whereas chemical assay for the same period reached 10 p.p.m. (Ware 1958). In general the results by bioassay were lower than by chemical assay except in some instances where the total BHC recovered by chemical assay was less than 0.5 p.p.m. It was also noted that after the termination of feeding, BHC was detectable by chemical assay longer than by bioassay. This was judged to be due in part to the more rapid rate of excretion of γBHC as compared with other more stable isomers of technical BHC undetectable by bioassay.

As testing proceeded it was noticed that fly mortalities in the Holstein standard curve were generally higher than the same concentrations in the Jersey milk. Therefore comparative standard curves were run with two series of milk samples, one varying in butter fat, the other varying in non-fat solids[3] (table 1). These data show that variations in butter fat result in significant variations in the toxicity of lindane applied in the milk. Variations in non-fat solids, mostly milk proteins, resulted in small differences in mortality. While the data from all concentrations of non-fat solids showed more than random variation by the Chi Square Test, this was due to the 9% non-fat solids milk exclusively. Results from the other skim milk samples did not vary significantly from each other.

What is the nature of the sparing action of butter fat on the toxicity of lindane? Laug (1948) analyzing BHC in rat tissues by means of house fly bioassay could detect amounts down to 0.2 p.p.m. in most tissues but "concentrations of gamma isomer lower than 10 p.p.m. in fat were considered outside the zone of reliability for bio-assay." He suggested that the fat "seemed to occlude gamma isomer and render it inaccessible to the fly." Returning to the data reported for milk we may explore the possibility that the flies limit their intake of the fat droplets in the milk so that only a portion of the fat is ingested. If so, then the higher the fat content of the milk, the less lindane would be present in the small amounts ingested. According to West (1951) the dimensions of the interbifid spaces on the house fly proboscis, namely 3 to 4 microns, limit the size of the food particles ingested during ordinary feeding; though much larger particles are occasionally taken up by "direct suction" feeding. The average fat particle size in fresh, whole, unhomogenized milk is 3 to 4 microns and in hand homogenized milk it is about 2 microns. Therefore, it appears that house flies cannot avoid ingesting the butter fat particles in freshly homogenized milk. (In milk which has "creamed" avoidance of fat by the flies is not only possible but readily observed; however the conditions of our experiment negate this possibility.)

If the flies cannot avoid ingesting the butter fat in homogenized milk they must take it freely in porportion to the amount present in the milk. To check this assumption flies were exposed 48 hours to untreated milk; one lot on skim milk, the other on 2.5% butter fat dairy homogenized milk. They were frozen, sexed, counted, dried under vacuum, and equal dry weights of female flies were extracted with ethyl ether. The ether-soluble extract from skim milk-fed flies weighed one-third less than the extract from the homogenized milk-fed flies. indicating that increased amounts of butter fat were ingested and retained by the flies on homogenized milk. It was frequently observed during this and other tests that flies on high butter fat milk egested much of the fat without digestion as accumulations of yellow butter fat were noted about the caudal ends of the flies.

Table 1.—Effect of varying content of butter fat and non-fat solids on the toxicity of lindane in milk fed to adult house flies.

PER CENT BUTTER FAT	PER CENT NON-FAT SOLIDS	ESTIMATED LC_{50} IN P.P.M. LINDANE
	Varying butter fat content	
0.0	9.0	0.34
2.5	9.1	0.64
4.8	9.2	0.93
6.5	9.3	1.4
	Varying non-fat solids	
3.8	9.0	0.98
3.8	12.0	1.15
3.8	15.0	1.18
3.8	17.0	1.25

This suggests that the reduced toxicity of lindane in the presence of butter fat is due to the reduced *chemical potential* of lindane under these conditions (Ferguson 1939). That is, for a given amount of lindane, the greater the amount of solvent butter fat, the lower will be the per cent saturation of lindane in the butter fat and the less will be its tendency to diffuse out of the butter fat and into the haemocoele and nerve tissues of the insect. A portion of the butter fat will pass through the alimentary canal and be egested, still retaining some fraction of its dissolved lindane intact. Finally, it should be pointed out that the reduced toxicity of lindane in the presence of butter fat poses no objection to the analysis of lindane by bioassay except as sensitivity is reduced, so long as the standard regression line is run on milk of the same type and butter fat content. The Toxicity Index (Sun 1957) will be the same even though the actual toxicities vary.

REFERENCES CITED

Ferguson, J. 1939. The use of chemical potentials as indices of toxicity. Proc. Royal Soc. (Series B) **127**: 387–403.

Laug, E. P. 1948. Tissue distribution of a toxicant following oral ingestion of the gamma isomer of benzene hexachloride by rats. Jour. Pharmacol. and Exptl. Therap. **93**: 277–81.

Sun, Y. P. 1953. Microbioassay of insecticides in milk by a feeding method. Jour. Econ. Ent. **46**: 927–30.

Sun, Y. P. 1957. Bioassay of Pesticide Residues; p. 449–96 in "Advances in Pest Control Research," Vol. I (R. L. Metcalf, Editor) Interscience Publ., New York.

Ware, G. W., Jr. 1958. Excretion of BHC in milk from dairy cows fed known amounts of BHC. Ohio Agric. Expt. Sta. Circ. In press.

West, L. S. 1951. "The Housefly" Comstock Publ. Co., Ithaca. 584 pp.

[3] Special milk samples supplied by Dr. W. J. Harper, Department of Dairy Technology, Ohio State University.

Tachinid Flies Reared from Lepidopterous Larvae in Arizona, 1957[1]

GEORGE D. BUTLER, JR., *University of Arizona Agricultural Experiment Station, Tucson*

During 1957, approximately 12,000 lepidopterous larvae from crop areas in Arizona were reared to determine the numbers and kinds of insect parasites present. Individual larvae were collected from cotton, alfalfa, corn and sorghum. One collection was also

[1] Arizona Agricultural Experiment Station Journal Series Paper No. 466. Accepted for publication May 12, 1958.

made from desert weeds 5 miles from the nearest agricultural area. The larvae were held in ½-pint waxed paper cups with transparent plastic lids and fed fresh alfalfa. The species of tachinid flies which emerged were determined by H. J. Reinhard.

From 125 records of tachinid flies recovered from these rearings, information was obtained on the common species present in crop areas in southern Arizona and their hosts. In addition, a summary of the distribution of the common species in Arizona is given as determined by 400 adult collection records. The adults were collected in the survey samples described by Butler & Werner (1957) and Werner & Butler (1957). Information on the hymenopterous parasites reared from the lepidopterous larvae in 1957 will be reported later.

Achaetoneura archippivora (WILLISTON).—This species is by far the most abundant tachinid fly parasite in Arizona crop areas and has the widest host range. The hosts from which it was reared and the number of collection records include: *Laphygma exigua* (Hbn.) (27), *Trichoplusia ni* (Hbn.) (18), *Laphygma frugiperda* (J. E. Smith) (7), *Heliothis* spp. (6), *Colias philodice eurytheme* Bdv. (3), *Loxostege similalis* (Guen.) (2), *Prodenia ornithogalli* Guen. (1), *Vanessa cardui* (L.) (1) and *Estigmene acrea* (Drury) (1). There were 70 rearing records obtained from the following crops and months: alfalfa (34), cotton (20), corn (7), sorghum (3) and weeds (6); April (1), May (2), June (15), July (26) and August (26).

A. archippivora is found throughout southern Arizona but has not been found in our samples from northern Arizona. It is most abundant in the summer and fall. Adults were collected during the winter but were very scarce during the spring.

Archytas marmoratus (TOWNSEND).—This species was reared from two collections of *Laphygma frugiperda* on corn from Tucson and Pomerene. One was made in June and the other in July. In 1931 specimens were reared from *L. frugiperda* collected in Tucson. Additional adults have been caught in two mountain areas near Tucson, Bear Canyon in the Catalina Mts. in November and Madera Canyon in the Santa Rita Mts. in September.

Blepharigena spinosula (BIGOT).—A single fly was reared from *Pseudaletia unipuncta* (Haw.) (det. E. L. Todd) collected on alfalfa at Amado in August. Adults have been collected in the southern half of the state with a single record from Navajo County. It is most abundant during June and July.

Chaetogaedia SP. NR. *analis* VAN DER WULP.—One specimen was reared from *Heliothis* spp. collected at Continental on corn in June. Adults have been collected in southeastern Arizona in Pima, Santa Cruz and Cochise Counties.

Eucelatoria armigera (COQUILETT).—This is primarily a parasite on *Heliothis* spp. Of 19 records, 18 were on *Heliothis* spp. and only one on *Trichoplusia ni*. Larvae feeding on corn (9), alfalfa (7), cotton (2) and weeds (1) were attacked. Rearings were made in May (2), June (6), July (4), and August (7). Although this species is distributed over the southern half of the state, it appears to be somewhat more abundant in the southeastern part.

Nemorilla floralis (FALLÉN).—One specimen was reared from *Loxostege similalis* which was on ragweed growing along the road east of Willcox. This species is widely distributed throughout western and central Arizona and the valleys and mountains of southeastern Arizona. It was found in alfalfa fields in the spring and fall.

Phorocera tachinomoides (TOWNSEND).—Found on a green looper on acacia at Sedona and on a skipper, *Lerodea eufala* Edw. (Det. W. D. Field) on alfalfa at Florence in August. This species is distributed over the entire state, most abundantly in the southern half, not only in the agricultural areas but in the mountain areas as well. Adults are most abundant during the spring and summer and have not been collected during the winter.

Plagiomima cognata ALDRICH.—Three rearings were made from *Heliothis* spp. swept from alfalfa at Yuma. Two were in July and one in August. Except for two adults, one from Cochise County and one from Pima County, the other six adult records are also from Yuma County.

Voria ruralis (FALLÉN).—This is the common tachinid parasite on *Trichoplusia ni*. There were 22 rearing records from this host, one from *Laphygma frugiperda* and one from a "green looper," on mesquite. Host larvae were obtained from cotton (12), and alfalfa (8) during May (3), June (4), July (7) and Aug. (10). *T. ni* was particularly severely infested by polyhedrosis virus and had a high mortality in the rearing containers so many additional parasite records were not obtained owing to the death of the host caused by disease. *V. ruralis* is commonly found in the crop areas of southern Arizona throughout the year. It is found only in the agricultural areas.

Zenillia hyphantriae (TOWNSEND).—This parasite was reared from *Loxostege similalis* on weeds along the highway east of Willcox in August. Adults have been taken in western, central and southeastern Arizona in April, May and July.

REFERENCES CITED

Butler, G. D., Jr., and F. G. Werner. 1957. The syrphid flies associated with Arizona crops. Arizona Agric. Expt. Sta. Tech. Bull. 132.

Werner, F. G., and G. D. Butler, Jr. 1957. The reduviids and nabids associated with Arizona crops. Arizona Agric. Expt. Sta. Tech. Bull. 133.

Endrin and Ryania for Control of Sugarcane Borer in Corn[1]

K. D. ARBUTHNOT, *Entomology Research Division, Agr. Res. Serv., U.S.D.A.*[2]

Insecticide tests for control of the sugarcane borer (*Diatraea saccharalis* (F.)) on corn were conducted at the Angleton substation of the Texas Agricultural Experiment Station in the summer of 1956. Little information on the seasonal history of the borer in this area was available to serve as a guide in timing the applications. Four replicates in a randomized block of plots four rows wide and 30 feet long were used in each of two tests. Three insecticide formulations were tested, dusts and granules of endrin, an insecticide highly toxic to the southwestern corn borer (Arbuthnot & Walton 1954), and a ryania dust recommended for control of the sugarcane borer in sugarcane (Ingram *et al.* 1951).

In the first test, five weekly applications were made on corn which was in the late-whorl stage when the first application was made on May 24. On this date 20% of the plants were infested with larvae in the second to sixth instars. (In larger corn in a nearby field freshly formed pupae were found.) All these forms were probably of the first generation. No eggs were found on plants in the area.

On July 10 ten plants were taken at random from the two center rows of each plot and dissected to determine the borer population (table 1). Pupal cases found were classified as first-generation, although some may have been second-generation forms since they were found in twice as many plants (40%) as the number infested by the first generation on May 24 (20%). There was no indication that endrin dust and granules affected the numbers of plants infested, but the percentage was lower for ryania. The endrin treatments reduced the first generation but were less effective than ryania. Both endrin treatments reduced the percentage of plants infested by the second generation, whereas all plants were infested in the ryania-treated plots. The borer populations were greatly reduced by both endrin treatments, but ryania was much less effective. Borers found in the ears included: in the untreated plots, all larval instars, pupae and pupal cases; in the endrin dust and ryania plots, all forms except first-instar larvae and pupal cases; and in the endrin granule

[1] Accepted for publication June 3, 1958.

[2] In cooperation with the Texas Agricultural Experiment Station. J. C. Smith, superintendent, and B. E. Jeter, Jr., agronomist of the substation staff, assisted in this study.

Table 1.—Effectiveness of five weekly applications of endrin and ryania against the sugarcane borer in early-planted corn. 1956.

Insecticide	Pounds per Acre	Per Cent of Plants Infested by Generation Shown			Per Cent of Ears Infested	Total Number of Borers in Ears	Per Cent Reduction of Borers	
		First	Second	Both			First Generation	Second Generation
Untreated[a]	—	40	100	100	85	33	—	—
Endrin:								
2.5% granules	40	38	40	70	15	9	42	95
1% dust	100	40	50	72	30	26	33	89
Ryania, 40% dust	62.5	25	100	100	52	28	67	42

[a] Untreated plants had 1.2 first-generation and 10.6 second-generation borers per plant.

Table 2.—Effectiveness of four applications of endrin and ryania against the sugarcane borer in late corn.[a]

Insecticide	Pounds per Acre	Stalks Broken	Plants Infested	Joints Bored	Ears Classed as Nubbins	Reduction		Increase in Yield
						Borers	Shanks Bored	
Untreated[b]	—	7	95	81	79	—	—	—
Endrin:								
2.5% granules	40	27	12	5	27	94	93	155
1% dust	100	16	75	17	36	74	57	160
Ryania, 40% dust	62.5	15	88	48	53	49	28	54

[a] Results are expressed in per cent.
[b] Untreated plants had 3.5 borers and 1.4 shanks bored per plant and a yield of 5 bushels per acre.

plots, only third-, fourth-, and sixth-instar larvae. Protection of the ears from infestation was best with the granules, followed by endrin dust and ryania. Endrin granules were outstanding in the reduction of borer populations in the ears. No yield data were obtained.

In a second test, on late-planted corn, the first of four applications was made on July 21. About 5% of the plants in this field harbored second-generation borers on July 10. Records taken on September 25 are summarized in table 2. Data on infestation, joints bored, borers, and shank damage were obtained by examining a random sample of 10 plants from each plot. Yield, nubbin, and stalk-breakage data were taken from the entire plot. The crop was severely affected by drought, and the stalks were weak, which may account for the higher stalk breakage associated with higher control of the borer, since the weight of an ear apparently caused the stalk to break. Endrin granules gave the best results in reducing plant infestation, joints bored, ear size (nubbins were ears less than one-half the average length), borer population, and shanks bored; and ryania was the least effective. Endrin granules and dust gave about equal yields and produced a threefold increase over the untreated check, whereas ryania gave a much smaller yield increase.

The rates and numbers of applications tested probably could not be used economically on most corn crops. However, the increases in yield resulting from borer control indicate the severity of damage that can be caused by the borer. More economical control measures, with lower dosages or fewer applications, or both, might be used profitably on corn having a higher potential yield than that used in the second test.

References Cited

Arbuthnot, K. D., and R. R. Walton. 1954. Insecticides for control of the southwestern cornborer. Jour. Econ. Ent. 47(4): 707.

Ingram, J. W., E. K. Bynum, Ralph Mathes, W. E. Haley, and L. J. Charpentier. 1951. Pests of sugarcane and their control. U. S. Dept. Agric. Circ. 878, 38 pp.

OBITUARIES

Henry James Franklin
1883-1958

Henry J. Franklin was born in Guilford, Vermont, February 10, 1883, and died at Buzzards Bay, Massachusetts on April 16, 1958. After the death of his father in Vermont, the family moved to Bernardston, Massachusetts, where he attended the public schools and prepared for college at Powers Institute. In 1899 he entered the Massachusetts Agricultural College in the class of 1903. He received his Ph.D. degree in entomology at Massachusetts Agricultural College in 1908.

While at college he showed his versatility by participating with success and distinction in many extracurricular activities. He won a prize for public speaking in his sophomore year, the first Flint (oratory) prize in his junior year, and was class orator at commencement. He was on his class yearbook board, the college senate, and played guard on the varsity football team for three years. His record in college, however, was chiefly characterized by serious and unremitting application to learning. He stood at the head of his class at graduation.

Dr. Franklin first came to study the cranberry insect situation during the summers of 1906 and 1907 when he was employed as an assistant by the Department of Entomology and stationed at Wareham, Massachusetts, in the heart of the Cape Cod cranberry district.

After receiving his Ph.D. degree in 1908, he served for a short time as assistant to the state entomologist of Minnesota. Late in 1909 Dr. Franklin came back to Massachusetts to organize the work and become head of the newly established Cranberry Station of the Massachusetts Agricultural Experiment Station at East Wareham, a position to which he gave the full benefit of his remarkable abilities and in which he served with faithfulness and devotion until his retirement in 1952. Under his leadership the Cranberry Station expanded to cover a wide range of research in many branches of cranberry culture—insect and disease control, bog management, varieties, weed control, and storage problems. One of his outstanding achievements was the study of weather affecting the cranberry industry, and particularly methods of forecasting the frost hazard on the bogs during the spring and fall months.

Dr. Franklin was the author of a long list of annual reports and bulletins and inspired many valuable contributions by his associates at the Station. His bulletins, *Cranberry Insects in Massachusetts*, *Weather in Cranberry Culture*, and *Cranberry Growing in Massachusetts*, have been called the cranberry growers' bible and have had a field of usefulness far beyond the boundaries of Massachusetts wherever cranberries are grown. He was the author of many publications on other types of insects, including his pioneer work on North American Thrips and his *Monograph of the Bumble Bees of the New World* is still a standard work on that group and won for him world-wide recognition.

He was elected to Phi Kappa Phi and Sigma Xi and was a member of the Entomological Society of America, the American Association for the Advancement of Science, the American Meteorological Society, and the Boston Society of Natural History.

In recognition of and appreciation for the help Dr. Franklin had rendered even beyond the official "call of duty," on his retirement in 1952, the cranberry growers of Massachusetts and other states raised a sum of money and built an addition to the Cranberry Station building which is known as the Doctor Franklin Room. Here he came almost daily, until failing health prevented, to work with his first interest, bumblebees, and to be among his friends, the cranberry growers and his colleagues at the Station.

In June 1953, after his retirement, the University of Massachusetts conferred on him an honorary Doctor of Science degree in recognition of his distinguished attainments.

Dr. Franklin married Esther M. Smith of Jamaica, New York, in 1918, who survives him, as do a brother and sister and several nephews.

In the years of his service at the Station, Dr. Franklin's work was a vital factor in the growth of the cranberry industry, not only in Massachusetts but throughout the country. This is witnessed by the fact that at the time of his retirement Massachusetts growers, from approximately the same acreage, were producing nearly double the yields which were being secured when Dr. Franklin first began his studies of cranberry growing problems.

Dr. Franklin was somewhat of a rugged individualist who seemed to prefer to work out his problems and draw his own conclusions rather than follow the easy path of accepting the ready-made opinions of others. While he was occasionally considered as rather conservative, it is significant that once he had solved a problem to his own satisfaction and had taken a firm stand, Dr. Franklin seldom if ever found it necessary to take a backward step. The cranberry growers of the Cape wore a well beaten pathway to his door and looked upon him as their refuge in times of trouble.

Dr. Franklin's record is an enduring monument, not only to his outstanding professional ability, but to his sterling character,

his unswerving integrity and unshakeable loyalty to his firm convictions. In his passing Entomology has lost a brilliant and outstanding member of the profession; those with whom he worked and for whose welfare he labored, a faithful ally; and those who were privileged to know him personally, a loyal and beloved friend.

W. E. TOMLINSON, JR.
A. I. BOURNE

O. S. Bare
1890-1958

Orlando S. Bare, Associate Professor of Entomology and Associate Entomologist of the Agricultural Experiment Station, University of Nebraska, Lincoln, died March 31, 1958, at the age of 67. He is survived by his wife, the former Aline Norris, son James, a daughter Joyce (Mrs. Donald Voils) and four grandsons.

Professor Bare was born December 15, 1890 near Clearwater, Nebraska. He received his B.S. degree in 1926 and his M.Sc. degree in 1929 from the University of Nebraska. He taught in the rural schools of Antelope County, Nebraska, from 1912–1915 and again from 1926–1928. During World War I he served overseas in Co. F, 109th Engineering Corps.

Mr. Bare was appointed Extension Entomologist in Nebraska in 1929. He served the state in that capacity until 1939 at which time he transferred to the teaching staff of the College of Agriculture as Assistant Professor. In 1942 he was promoted to the rank of Associate Professor.

"Dan" Bare, as he was known to his close associates, was extension entomologist during the 1930's and effectively "generalled" the "war" against the heavy outbreaks of grasshoppers which followed a series of drouth years. He became widely known and respected throughout Nebraska for his sound practical approaches to the solution of all pest problems. He was an authority on bees both academically and from first-hand experience for he kept a few hives of his own and managed the Department of Entomology colonies maintained for demonstration and classroom purposes. He was a member of the American Beekeeping Federation, the Entomological Society of America, Kansas Entomological Society, Sigma Xi, Alpha Zeta, Gamma Sigma Delta and Phi Sigma.

Although Mr. Bare's talents were exhibited primarily in the field of extension and teaching, he retained an interest in the taxonomy of ants and was ever ready to lend a hand with research projects of other staff members, especially during the summer months. Most of his writing and publishing took the form of numerous extension circulars and bulletins.

Mr. Bare possessed a keen sense of humor, which enabled him to get much enjoyment out of life. He followed sports, liked to fish and bowl and played a good hand of cribbage. One of his principal hobbies was gardening, which he pursued to the point of becoming a local authority in the art. His friends were many, including neighborhood youngsters who liked to go "talk with Mr. Bare."

O. S. Bare

O. S. Bare was very much a part of Nebraska entomology throughout his 29 years of loyal and devoted service. His counsel and valued friendship will be sorely missed and long remembered by his students and those of us privileged to be associated with him professionally.

ROSCOE E. HILL

Easy-to-apply Shell Chemical insecticides give dependable control.

Gone are the "loss and buggy" days!

The days of farmers' complete crop losses to "bugs" are rapidly disappearing and will soon be only a memory like the horse and buggy. Today's insecticides give a decided advantage to farmers. With dependable insect control, they now market a much larger share of harvests.

To discover newer and better means of protecting crops, Shell Chemical Corporation researchers work with State and Federal experimental stations, independent researchers, extension workers and growers.

As a result of intensive experimentation, Shell developed aldrin, dieldrin, endrin and Phosdrin® insecticides; D-D® and Nemagon® soil fumigants; and Allyl Alcohol weed seed killer.

Other pesticides being developed in experimental installations throughout the country promise even greater success for agriculture—and a bleak future for insect pests!

Technical information on Shell Chemical Corporation pesticides is available. Write to:

SHELL CHEMICAL CORPORATION

AGRICULTURAL CHEMICAL SALES DIVISION

460 PARK AVENUE, NEW YORK 22, NEW YORK

SHELL